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# EFFECT OF STORAGE ON LEACHING OF MINERALS AND NITROGEN FROM ASPARAGUS AND PEAS DURING COOKING[1]


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## INTRODUCTION

FRESH GREEN ASPARAGUS and peas are recognized as highly nutritious vegetables and, when properly cooked, good sources of minerals. California produces more than half of all the asparagus in this country for both canning and marketing, and a large proportion of peas for marketing fresh, for canning, and for quick freezing. Asparagus and peas produced here are stored for various periods before reaching the consumer. Several days usually elapse between the harvest of the vegetables and their consumption. As has been known for some time, asparagus and peas undergo significant changes during storage. Bisson, Jones, and Robbins (*1*)[5] report changes in crude fiber, sugar, dry matter, and weight of asparagus stored at various temperatures for various periods.

Bisson, Jones, and Allinger (*2*) report changes in peas stored at 25° C. When the fruit was stored unshelled, there was a rapid translocation of material from pods to peas, as indicated by the increasing weight of the


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[5] Italic numbers in parentheses refer to "Literature Cited," at the end of this paper.

dry material. These authors report also that the elements nitrogen, carbon, phosphorus, and magnesium were translocated in determinable amounts to the peas.

The experiments here reported were begun primarily to determine the influence of different storage periods upon the amounts of minerals and nitrogen leached from asparagus and peas during cooking and the changes, if any, in crude-fiber content. The minerals studied were magnesium, calcium, phosphorus, copper, and iron. In addition, total nitrogen and crude fiber were determined.

Considerable work has been done on the mineral losses sustained when vegetables are cooked (*3, 4, 5, 6*). But hitherto, most of the work reported on vegetable cookery has been done on typical market vegetables of uncertain variety and source. As investigators have shown (*7, 8, 9, 10, 11, 12, 13, 14, 15*), the quality of asparagus and peas is influenced by cultural practices, fertilizer treatment, the nature of the soil, seasonal conditions, the stage of development at harvesting, and the methods of handling and storage employed thereafter. In the experiments herein reported, not only the variety but also the cultural practices were controlled, together with the maturity and freshness of the vegetables.

As far as we are aware, no work has been done on storage as affecting the amounts of mineral constituents and nitrogen leached from asparagus and peas of a known variety and source during cooking. Since these vegetables are always eaten cooked, it is important, from the standpoint of human nutrition, to determine the exact mineral losses, if any, during the cooking processes. It is important also from a consumer standpoint to determine the effect of storage on mineral losses sustained in cooking because of the delay that usually occurs between harvest of market vegetables and their consumption.

## MATERIAL AND METHODS

*Preparation of Asparagus Samples.*—The asparagus used in these experiments was Palmetto. All the asparagus was uniformly collected before 6:45 A.M. on April 14, 1936. The work of preparing the samples for storage was carried out in the cold-storage room in order to keep the asparagus as fresh as possible. It took approximately 4 hours.

Samples were composed of 40 spears selected in the field as representative for uniformity of size, height, and cleanliness. Two size grades were used: ⅜ to ½ inch and ½ to ⅝ inch. (The diameter was measured 3 inches from the tip.) After grading, the spears were bunched and trimmed to a length of 5½ inches. Each bunch was weighed immediately to a tenth of a gram; the weight of each was approximately 400 grams.

After the sample had been weighed, the spears were washed in distilled water to remove sand adhering to their surfaces. The amount of sand thus washed out was saved to determine the amount of correction to be applied to the weight of the fresh asparagus. As the average weight of the sand was less than 0.10 gram, it was considered negligible, and no correction was applied to the original weight. After having been washed, the samples were placed in 1-liter beakers with the butt ends resting in about ⅓ inch of distilled water. This corresponds more or less with commercial practice, for asparagus is packed with the butts resting on moist moss or some other absorbent.

All samples were then taken to an unventilated storage room where the temperature over the 10-day storage period averaged 35 ± 2° F. The storage periods were for 0, 24, 48, 96, 168, and 240 hours. These intervals correspond to time required to transport asparagus to different sections of the country. Six samples chosen at random the first day of the experiment comprised the master sample, which was not cooked. Twelve samples were removed from the storage room at the end of each period. Four of these were not cooked, and eight were cooked.

*Preparation of Pea Samples.*—Marketable peas, Giant Stride variety, mechanically shelled from Fancy and U. S. No. 1 grades, were used in this experiment. The peas were picked, shelled, and weighed on the same day, May 18, 1936. The shelling and the preparation of samples for storage were carried out in the cold-storage room in order to keep the peas as fresh as possible. After shelling, the peas retained by screens 5, 6, and 7 (from about 22/64 to 28/64 inches in diameter) were thoroughly mixed so as to yield uniform samples. Each sample was weighed to a tenth of a gram; the weight of each was approximately 400 grams. Each was then put in a liter beaker. Fifty-eight samples were used in analyses. All samples were stored uncovered in a storage room for the required time. Moist moss was kept on the floor of the storage room. The average temperature of the storage room was 34 ± 2° F, and the storage periods were the same as for asparagus. Four unstored, uncooked samples comprised the master sample.

The 58 samples were divided among the storage periods and methods of treatment as follows:

| Storage period | Number uncooked | Number boiled | Number steamed |
|---|---|---|---|
| 0 | 4 | 4 | 4 |
| 24 | 3 | 3 | 4 |
| 48 | 3 | 4 | 3 |
| 96 | 3 | 3 | 3 |
| 168 | 3 | 3 | 3 |
| 240 | 2 | 3 | 3 |

Originally it was planned to have as many samples as in the asparagus work, but unfortunately there were only enough peas to make 58 samples of 400 grams and 6 samples of 50 peas each.

The 6 samples of 50 peas each were taken at random, and puncture tests were performed on them to determine changes in toughness of the pericarp, probably associated with changes in the crude fiber. The puncture tests were made on 50 seeds the day the peas were picked and at each storage period thereafter by means of a standard apparatus loaned by the United States Department of Agriculture Bureau of Plant Industry. A reading of one unit on the puncture-test scale corresponds to a spring depression of 10 grams. The needle used was blunt, with a diameter of 0.02 cm and an area of 0.0003 sq. cm.

*Methods of Cooking.*—The procedure was standardized in an endeavor to reduce experimental errors to a minimum. Two different methods of cookery were used throughout the experiments: steaming and boiling. In cooking, the aim was to make the vegetable look attractive and taste good. Aluminum vessels were selected for boiling and steaming. Distilled water was used: 250 cc for steaming and 2,500 cc for boiling. All cooking was done on 1,000-watt electric hot plates. In every case, the water was first brought to a boil before the vegetable was added.

The samples of asparagus to be cooked were gently washed in distilled water to remove adhering sand, care being taken not to break off the scalelike leaves and spear heads.

In the boiling method the asparagus spears were dropped into the rapidly bubbling water, and the time for the cooking, counted from the instant the water came back to the boiling stage, was 20 minutes.

In the steaming method the asparagus spears were laid in layers across a perforated aluminum disk, and the time was counted as soon as the last spear had been placed. Quadruplicate samples of asparagus of each storage period were cooked by each method.

The samples of peas were emptied directly into the cooking vessel from the liter beaker in which they were stored.

*Drying of Samples and Evaporation of Extract.*—At the close of the cooking period, the samples of asparagus were carefully lifted from the cooking water and from the aluminum disk, drained for 1 minute, and placed on trays. The water was then filtered to remove the small particles of asparagus broken off from the spears during cooking and the sand that had lodged underneath the scalelike leaves and in the spear heads. The cooking water from each sample was evaporated, and the residue partially dried over a water bath. The partially dried residues were then transferred to an electric oven and dried for approximately 3 days at

149° F, or until the variation of two consecutive weighings was not greater than 0.05 gram. They were then stored for later analysis.

The cooked and uncooked samples of asparagus were placed crosswise on wire trays especially made for this experiment and were dried in a homemade drier for 2 days at about 149° F. A vacuum motor, circulating the hot air through the drier, enabled the samples to dry as uniformly as possible. The partially dried samples were then transferred to an electric oven, and the drying was continued at the same temperature until two consecutive weighings agreed within 0.20 gram. The dried samples were then broken up by hand and were ground in an iron mill so that they would pass through a 40-mesh screen, except the master samples, which were ground by hand in a porcelain mortar. The ground samples were redried at 167° F to constant weight, after which they were placed in bottles, sealed airtight with paraffin, and stored for later analysis.

The drying of the pea samples, the evaporation of extracts, and the grinding of the dried material were the same as that described for the asparagus samples, except for slight differences in handling because of the difference in type of vegetable.

*Analytical Methods.*—The ground samples were ashed and made into solution with dilute nitric acid.

The dried cooking-water residue of each cooked sample was dissolved by adding 5 cc of sodium citrate (1 cc equivalent to 5 mg of sodium) and enough hot water to make a uniform solution. This solution was then evaporated to dryness over a steam bath in the electric muffle. Next, the ash was dissolved in dilute nitric acid, evaporated to dryness, and baked in an electric oven for 2 hours at 230° F to render the silica insoluble. The dried residue was dissolved with 10 cc of 6*N* nitric acid and 20 cc of water. The solution was filtered into a 100-cc volumetric flask, and volume made to mark. A 10-cc aliquot was removed for the phosphorus determination. The remainder was used for determining calcium, magnesium, copper, and iron.

The procedure outlined in the official methods of analysis (*16*) was used in determining calcium and magnesium.

Phosphorus was determined essentially according to the official methods (*16*), with this modification: The phosphorus was precipitated as ammonium phosphomolybdate and digested for 30 minutes at a temperature of 113° to 122° F. The precipitate was then filtered on an asbestos mat, washed with boiled water, dissolved in excess standard sodium hydroxide, and boiled for 30 minutes. After cooling, the excess alkali was titrated with standard hydrochloric acid, phenolphthalein indicator being used. This modification gives a sharper end point in the final titration.

Copper and iron were determined from the filtrate after calcium and magnesium had been removed. The citrates and oxalates, which interfere with the determination of iron, were decomposed before the iron and copper determinations were made. Iron was determined colorimetrically by the potassium thiocyanate method (*16*). Elvehjem and Lindlow's (*17*) method was used in determining copper.

Total nitrogen was determined according to the Kjeldahl, Gunning, Arnold method (*16*), except that selenium was used to hasten the digestion process.

The method used for determining crude fiber was that given in the official method of analysis (*16*).

## RESULTS WITH ASPARAGUS

*Edible Quality of Steamed and Boiled Asparagus.*—One of the most important aims in cooking a vegetable is to prepare an appetizing product of highest edible quality. Provided the harvesting is done at the proper time and the product stored at a suitable temperature, the most important factor influencing edible quality is the method of cooking. Color, flavor, and palatability influence edible quality. Unless a vegetable looks appetizing and tastes good, it is not readily eaten. In the preliminary tests, eight persons carefully compared steamed and boiled asparagus with respect to color, flavor, and palatability.

The color was altered during both processes of cooking. The bright green of the fresh material was changed to a darker green. When the cooked products were allowed to stand in air, the tips became much darker, probably because tannins and related substances were oxidized. No color differences could be distinguished between the steamed and the boiled product.

A pronounced characteristic asparagus flavor was common to both lots. The tips were mild with nutlike or meatlike flavor. There were no differences in sweetness. When cooked too long, asparagus develops an astringency or bitterness; but no such flavor could be detected in either the boiled or the steamed product. The spears in both lots were tender and made an agreeable food throughout their entire length. Although no differences in color, flavor, and palatability were noted between steamed and boiled asparagus, differences in mineral content were found when the respective products were analyzed.

*Composition of Asparagus Used in This Study and Uniformity of Samples.*—The partial composition of the master sample for the constituents studied in this experiment was as follows:

| Constituent | Milligrams in 400 grams of fresh asparagus |
|---|---|
| Copper | 0.76 |
| Iron | 3.60 |
| Calcium | 61.00 |
| Magnesium | 84.55 |
| Phosphorus | 326 |
| Nitrogen | 2,050 |
| Crude fiber | 2,810 |

In order to collate results, it was important to know whether or not the samples were uniform. Even though they had been selected carefully as to size, length, and color of spear, their uniformity could be established only by chemical analyses. Phosphorus was selected as the reference element. If the samples were uniform, then the amount of phosphorus leached during cooking plus the amount remaining in the residue should

TABLE 1

UNIFORMITY OF ASPARAGUS SAMPLES, EXPRESSED AS TOTAL PHOSPHORUS
(Storage temperature 35° F)

| Storage period | Total phosphorus | | | | | |
|---|---|---|---|---|---|---|
| | Master sample | | Boiled asparagus (residue+extract*) | | Steamed asparagus (residue+extract*) | |
| | Mean† | $s$‡ | Mean† | $s$‡ | Mean† | $s$‡ |
| *hours* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* |
| 0 | 326 | 4 | 329 | 12 | 349 | 13 |
| 24 | .... | .. | 328 | 6 | 320 | 12 |
| 48 | .... | .. | 330 | 9 | 320 | 10 |
| 96 | .... | .. | 337 | 5 | 332 | 8 |
| 168 | .... | .. | 324 | 7 | 325 | 8 |
| 240 | .... | .. | 326 | 7 | 322 | 7 |

* Cooking waters from boiling and steaming.

† The master sample consisted of six bunches of forty spears each; four such bunches were used for each cooking method at each storage interval.

‡ $s$=Estimated standard deviation = $\sqrt{\frac{\Sigma d^2}{n-1}}$.

equal the amount of phosphorus in the master sample. All samples of the steamed and boiled asparagus and their respective extracts (the cooking waters from steaming and boiling) were analyzed for phosphorus. The results are given in table 1. At each storage period, the total weight of phosphorus of both boiled and steamed asparagus was within about 2 per cent of that found in the master sample. Such consistent results show that the samples were uniform. The total nitrogen and crude-fiber analyses (discussed later in the paper) of the uncooked samples at the various storage periods further substantiate this uniformity.

*Effect of Storage on Minerals Leached from Asparagus during Cooking.*—The amounts of total solids in the asparagus extracts are represented graphically in figure 1. The extracts from the boiled asparagus contained approximately four times as much solid material as the extracts from the steamed. With both methods of cooking, the total solids

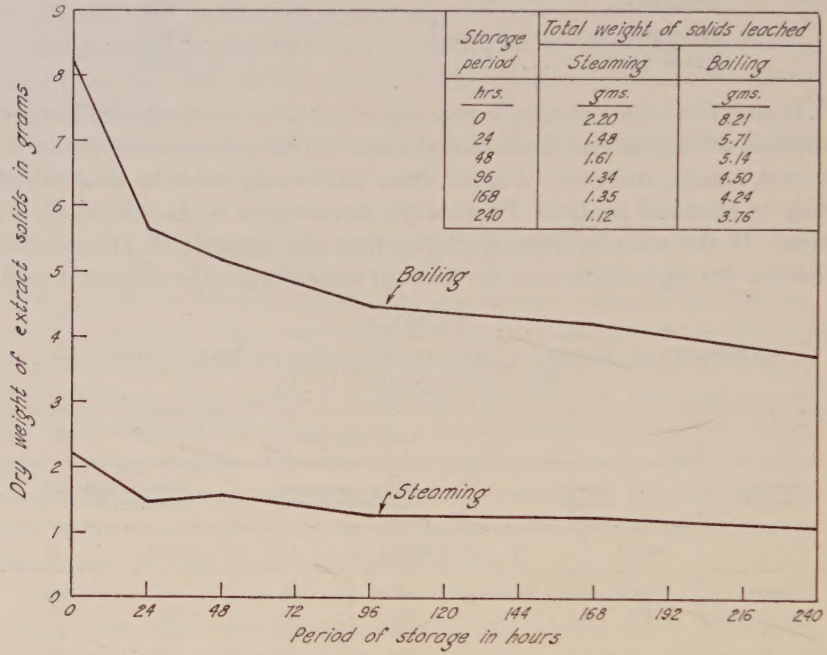


Fig. 1.—Total solids leached from asparagus spears during cooking (on 400-gram fresh basis).

in the extracts were greater when cooked immediately after trimming than when cooked after storage. The leaching of total solids decreased rapidly during the first 48 hours. After that time, the decrease was gradual.

Total solids of each extract for each storage period were analyzed for magnesium, calcium, phosphorus, iron, and copper. The results of the analyses for the first three of these elements appear in table 2. At each storage period, more magnesium, more calcium, and more phosphorus were leached out during boiling than during steaming. Steamed asparagus, therefore, contains more of these minerals than boiled asparagus. Judging from the wide difference in the amount of each mineral leached during boiling and steaming, the rate of leaching is dependent not only on the particular element but also on the amount of water coming in contact with and passing through the vegetable.

TABLE 2

MINERALS LEACHED FROM ASPARAGUS DURING TWO DIFFERENT METHODS OF COOKING, ON 400-GRAM FRESH-WEIGHT BASIS
(Storage temperature 35° F)

| Storage period | Magnesium* | | | | Calcium† | | | | Phosphorus‡ | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Boiling | | Steaming | | Boiling | | Steaming | | Boiling | | Steaming | |
| | Mean | $s$¶ | Mean | $s$ | Mean | $s$ | Mean | $s$ | Mean | $s$ | Mean | $s$ |
| *hours* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* |
| 0 | 32.84 | 1.72 | 10.06 | 2.32 | 17.20 | 1.70 | 4.76 | 0.89 | 67.59 | 2.84 | 18.43 | 3.40 |
| 24 | 26.71 | 2.31 | 7.04 | 0.91 | 13.45 | 1.25 | 3.91 | 0.53 | 50.20 | 3.21 | 13.23 | 1.51 |
| 48 | 23.38 | 0.92 | 6.75 | 0.94 | 12.00 | 0.76 | 3.99 | 0.38 | 43.44 | 1.27 | 14.10 | 1.48 |
| 96 | 21.46 | 2.34 | 6.79 | 0.91 | 11.05 | 0.31 | 3.78 | 0.20 | 41.01 | 2.80 | 13.15 | 0.86 |
| 168 | 20.41 | 0.02 | 7.63 | 0.69 | 10.30 | 0.75 | 3.50 | 0.65 | 39.27 | 2.00 | 13.58 | 1.09 |
| 240 | 19.22 | 0.42 | 6.18 | 0.58 | 7.94 | 1.40 | 3.26 | 0.24 | 40.16 | 0.47 | 12.25 | 1.37 |

* Master sample, $M$=84.55, $s$=3.27.
† Master sample, $M$=61.00, $s$=7.02.
‡ Master sample, $M$=326.10, $s$=4.18.
¶ $s$=Estimated standard deviation.

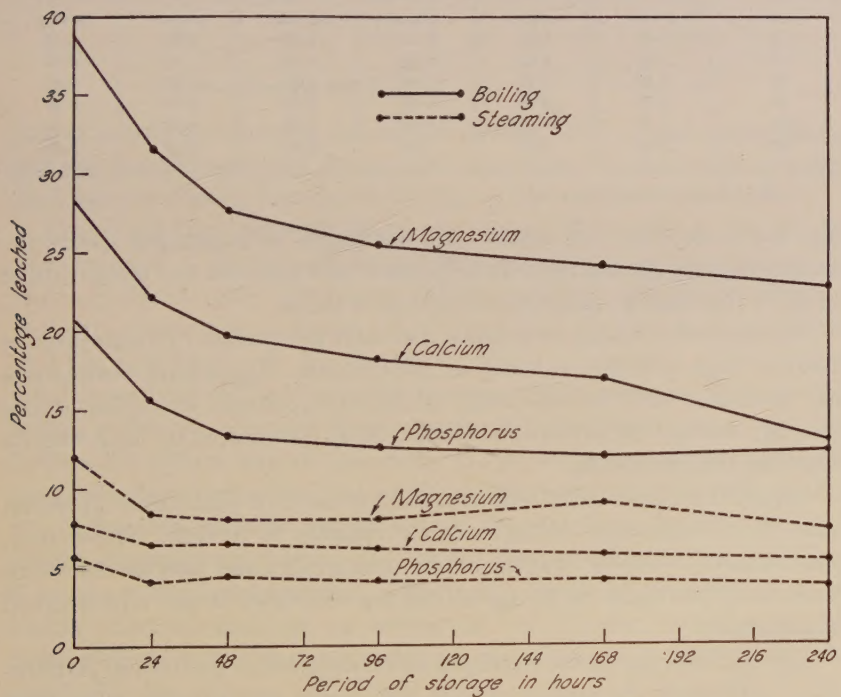


Fig. 2.—Percentage of minerals leached from asparagus during two different methods of cooking.

The percentages of magnesium, calcium, and phosphorus leached from boiled and from steamed asparagus are plotted in figure 2. With the boiled asparagus, the amount of each mineral leached decreased rapidly during the first 48 hours of storage. After that time, the rate of decrease in leaching was gradual. With the steamed asparagus, the rate of decrease in leaching was most rapid during the first 24 hours and then practically constant. This high initial decrease probably results from a rapid transference of the elements into less-soluble organic forms. Bisson

TABLE 3

TOTAL NITROGEN IN UNCOOKED AND COOKED ASPARAGUS AT DIFFERENT STORAGE PERIODS, ON 400-GRAM FRESH-WEIGHT BASIS

(Storage temperature 35° F)

| Storage period | Uncooked samples | | Cooked samples | | | |
|---|---|---|---|---|---|---|
| | | | Boiled | | Steamed | |
| | Mean | $s$* | Mean | $s$* | Mean | $s$* |
| *hours* | *grams* | *grams* | *grams* | *grams* | *grams* | *grams* |
| 0 | 2.05 | 0.03 | 1.76 | 0.05 | 2.08 | 0.05 |
| 24 | 2.01 | 0.02 | 1.84 | 0.02 | 1.95 | 0.05 |
| 48 | 2.05 | 0.10 | 1.89 | 0.06 | 2.05 | 0.01 |
| 96 | 2.10 | 0.09 | 1.98 | 0.06 | 2.08 | 0.03 |
| 168 | 2.10 | 0.06 | 1.96 | 0.05 | 2.10 | 0.06 |
| 240 | 2.04 | 0.03 | 1.90 | 0.07 | 2.04 | 0.06 |

* $s$=Estimated standard deviation.

and his co-workers (*1*) report that the length of asparagus spears increases rapidly during the first 48 hours of storage even at a temperature of 33° F, but that its subsequent increase is slight.

During both methods of cooking, a greater percentage of magnesium is leached than of either calcium or phosphorus. Magnesium compounds are generally more soluble than calcium compounds. Phosphorus was pobably held by the protein, which is readily coagulated by heat, and its leaching thus inhibited.

Copper and iron were found in the extracts in such small amounts that the results were not sufficiently reliable to include. These analyses do show, however, that the amounts of copper and iron leached were practically the same—0.10 mg to 0.20 mg—for both boiled and steamed asparagus.

*Effect of Storage on the Nitrogen and Crude-Fiber Content of Asparagus.*—Although the master sample was tested for nitrates, none were detected. The observation of Culpepper and Moon (*18*) that nitrates are absent in all young shoots and are present only in small amounts in shoots

72 inches long, explains this finding; all the asparagus shoots used in this study had been cut to a length of 5½ inches.

The results of the analyses for total nitrogen in the uncooked and cooked asparagus at different storage periods are presented in table 3. The total nitrogen of the uncooked samples at the various storage periods did not vary appreciably. Less nitrogen was found in the boiled asparagus than in the steamed. Obviously, some nitrogen was leached out. Amino nitrogen, which, according to Culpepper and Moon (*18*), is fairly equally distributed throughout the length of the spear, is soluble and

TABLE 4

CRUDE-FIBER CONTENT OF UNCOOKED ASPARAGUS, ON 400-GRAM FRESH-WEIGHT BASIS

| Storage period | Crude-fiber content | | Storage period | Crude-fiber content | |
|---|---|---|---|---|---|
| | Mean | $s$* | | Mean | $s$* |
| *hours* | *grams* | *grams* | *hours* | *grams* | *grams* |
| 0 | 2.81 | 0.07 | 96 | 2.86 | 0.06 |
| 24 | 2.78 | 0.02 | 168 | 2.83 | 0.07 |
| 48 | 2.80 | 0.09 | 240 | 2.80 | 0.08 |

* $s$=Estimated standard deviation.

was evidently leached out during the boiling. Little if any amino nitrogen was leached out from the steamed asparagus; the total nitrogen content was practically identical with that of the uncooked product.

The crude-fiber determinations are presented in table 4. The crude-fiber content of the uncooked asparagus at various storage periods did not vary significantly. The master sample was found to contain only 0.70 per cent crude fiber—a figure much lower than that reported by Bisson and his co-workers (*1*) for the same variety (Palmetto) grown under similar conditions. Their samples, however, were composed of shoots 8½ inches in length, whereas those used in this experiment were only 5½ inches. The spears used in this study, furthermore, were green from the tip to the basal segment; and in the preliminary cookery tests, shoots 5½ inches long made an agreeable food product throughout their entire length. Culpepper and Moon (*18*) report that in stalks 8 inches long, 1 to 2 inches of the basal segment is distinctly too tough and fibrous to be highly palatable and that an additional 1 or 2 inches contains fiber in noticeable amounts but not in sufficient abundance to make the material unsuitable for eating.

## RESULTS WITH PEAS

*Composition of Peas Used in This Study and Uniformity of Samples.*—The partial composition of the master sample for constituents studied in this experiment was as follows:

| Constituent | Milligrams in 400 grams of fresh peas |
|---|---|
| Magnesium | 118.20 |
| Calcium | 35.45 |
| Phosphorus | 517.40 |
| Iron | 6.71 |
| Copper | 3.53 |
| Nitrogen | 4,380.00 |
| Crude fiber | 6,280.00 |

To show the effect of storage on the leaching of minerals, each sample cooked must contain as much of a given constituent as any other sample; otherwise, the picture will be distorted. Even though sampling is done carefully to insure uniformity, a chemical analysis is necessary to establish this point. As with asparagus, phosphorus was chosen as the refer-

TABLE 5

Uniformity of Pea Samples, Expressed as Total Phosphorus
(Storage temperature 34° F)

| Storage period | Total phosphorus | | | | | |
|---|---|---|---|---|---|---|
| | Master sample | | Boiled peas (residue+extract*) | | Steamed peas (residue+extract) | |
| | Mean | s† | Mean | s | Mean | s |
| *hours* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* |
| 0 | 517 | 12 | 494 | 6 | 513 | 10 |
| 24 | .... | .. | 507 | 10 | 516 | 15 |
| 48 | .... | .. | 494 | 10 | 512 | 9 |
| 96 | .... | .. | 493 | 18 | 525 | 14 |
| 168 | .... | .. | 494 | 15 | 501 | 8 |
| 240 | .... | .. | 493 | 13 | 505 | 2 |

* Cooking waters from boiling and steaming.
† $s$ = Estimated standard deviation.

ence element. If the samples are uniform, then the phosphorus leached out during cooking plus the amount remaining in the peas should be constant or equal to the amount found in the master sample. The phosphorus found in the extracts (cooking water) has been added to the amount remaining in the cooked peas (residues), with the results shown in table 5.

The results indicate that we were successful in obtaining uniform samples: the total phosphorus was within an average of 3 per cent of the

total phosphorus found in the master sample. Further evidence indicating the uniformity of samples is that of the total nitrogen found in the uncooked samples. A glance at table 7 (p. 309) will prove this point, even though the value of the total nitrogen for the initial storage period is a little higher than the others.

*Effect of Storage on Minerals Leached from Peas during Cooking.*— The average weights of total solids leached at each storage period during both methods of cooking are graphically shown in figure 3. Approxi-

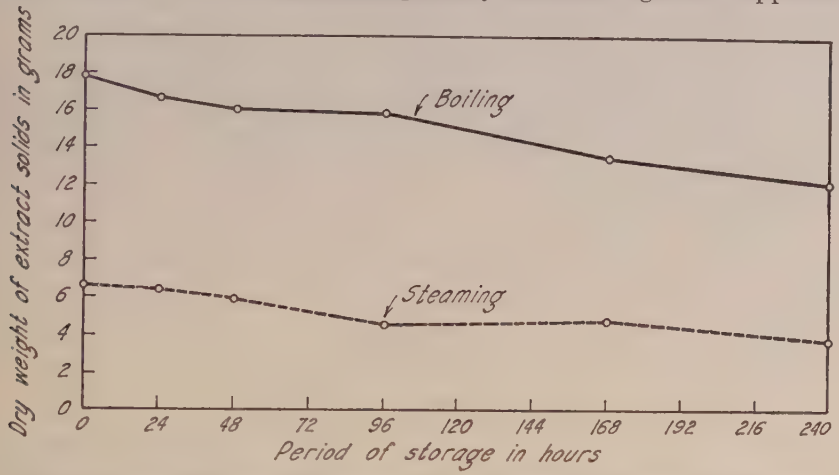


Fig. 3.—Total solids leached from peas during cooking (on 400-gram fresh basis).

mately three times as much solid material was leached from boiled peas as from steamed—a somewhat smaller difference than in asparagus. The amount of total solids leached by either method of cooking was between two and three times as much for peas as for asparagus.

In contrast with asparagus, in which the amount of leaching decreased rapidly during the first 48 hours of storage, the amount of solids leached from peas decreased gradually at each successive storage period. The rate of decrease of solids is practically the same for both methods of cooking; evidently storage affects the solids to the same degree for both.

The calcium, magnesium, and phosphorus leached by both cooking methods are recorded in milligrams per sample in table 6 and shown by percentage leached in figure 4. In every case, except for calcium at the 24-hour and the 48-hour storage period, a higher percentage of the mineral constituents was leached by boiling than by steaming.

A higher percentage of magnesium was leached out than of phosphorus or calcium, both by boiling and by steaming. In contrast with asparagus, where the order of decreasing percentage leached with both

TABLE 6

Minerals Leached from Peas during Two Different Methods of Cooking, on 400-Gram Fresh-Weight Basis

(Storage temperature 34° F)

| Storage period | Magnesium* | | | | Calcium† | | | | Phosphorus‡ | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Boiling | | Steaming | | Boiling | | Steaming | | Boiling | | Steaming | |
| | Mean | s¶ | Mean | s | Mean | s | Mean | s | Mean | s | Mean | s |
| *hours* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* | *mg* |
| 0 | 39.21 | 1.91 | 18.23 | 0.48 | 4.97 | 0.51 | 4.32 | 0.25 | 112.60 | 7.54 | 47.90 | 0.90 |
| 24 | 37.95 | 2.90 | 20.07 | 1.37 | 4.12 | 0.45 | 4.17 | 0.28 | 111.50 | 9.12 | 48.51 | 3.73 |
| 48 | 36.88 | 1.24 | 19.04 | 1.52 | 3.87 | 0.31 | 4.10 | 0.12 | 109.90 | 8.41 | 46.25 | 3.28 |
| 96 | 38.52 | 2.89 | 15.93 | 0.93 | 4.64 | 0.51 | 4.35 | 0.41 | 111.60 | 7.50 | 38.64 | 1.64 |
| 168 | 36.07 | 1.78 | 16.87 | 0.38 | 4.74 | 0.64 | 3.81 | 0.44 | 102.60 | 6.32 | 41.20 | 2.33 |
| 240 | 35.33 | 2.14 | 14.18 | 0.50 | 4.18 | 0.38 | 3.50 | 0.61 | 98.50 | 5.72 | 34.19 | 2.02 |

* Master sample, $M=118.2$ mg, $s=5.69$ mg.
† Master sample, $M=35.45$ mg, $s=1.21$ mg.
‡ Master sample, $M=517.4$ mg, $s=12.30$ mg.
¶ $s$ = Estimated standard deviation.

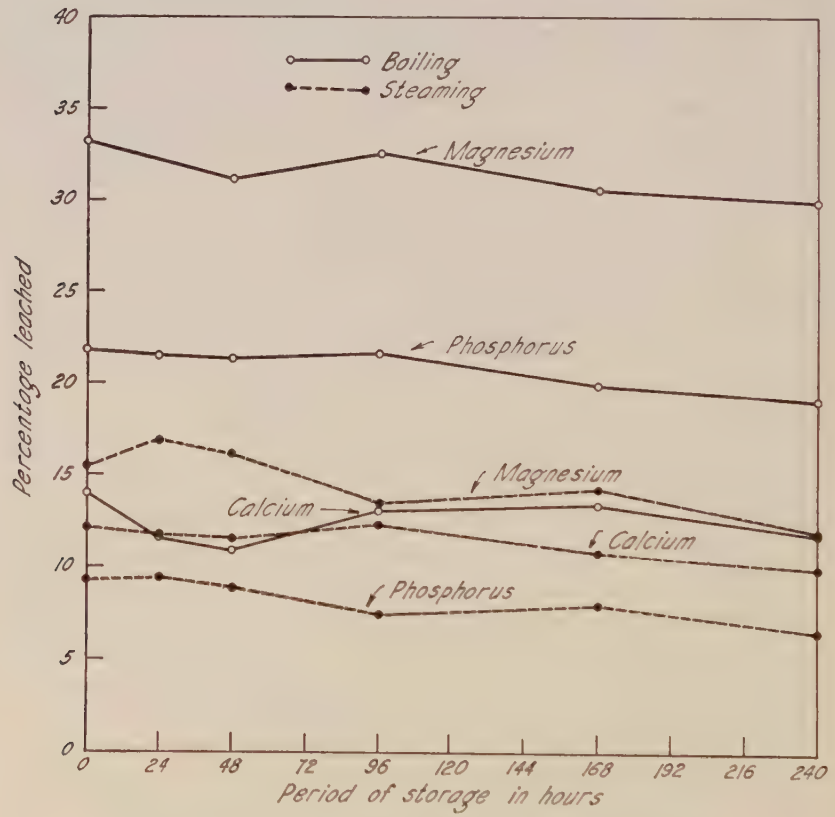


Fig. 4.—Percentage of minerals leached from peas during two different methods of cooking.

methods of cooking was magnesium, calcium, and phosphorus, that with peas was magnesium, phosphorus, and calcium.

Storage slightly decreased the percentages of magnesium, phosphorus, and calcium leached, but the differences with calcium are not significant.

Approximately twice as much magnesium and phosphorus were leached out by boiling as by steaming. Calcium, however, was leached only slightly more by the boiling. At the 24-hour and 48-hour periods, in fact, the percentages of calcium leached was greater for the steam-cooked

TABLE 7

TOTAL NITROGEN IN UNCOOKED AND COOKED PEAS AT DIFFERENT PERIODS, ON 400-GRAM FRESH-WEIGHT BASIS

(Storage temperature 34° F)

| Storage period | Uncooked samples | | Cooked samples | | | |
|---|---|---|---|---|---|---|
| | | | Boiled | | Steamed | |
| | Mean | $s$* | Mean | $s$* | Mean | $s$* |
| *hours* | *grams* | *grams* | *grams* | *grams* | *grams* | *grams* |
| 0 | 4.38 | 0.09 | 3.45 | 0.16 | 3.84 | 0.09 |
| 24 | 4.12 | 0.23 | 3.52 | 0.27 | 3.94 | 0.19 |
| 48 | 4.08 | 0.05 | 3.49 | 0.16 | 3.93 | 0.14 |
| 96 | 4.18 | 0.08 | 3.47 | 0.23 | 4.11 | 0.11 |
| 168 | 4.24 | 0.10 | 3.56 | 0.23 | 3.85 | 0.09 |
| 240 | 4.10 | 0.06 | 3.56 | 0.21 | 3.95 | 0.07 |

* $s$=Estimated standard deviation.

peas. The greater irregularity of the calcium curve for boiled peas is no doubt due to the combined effect of slight nonuniformity of samples and the greater error involved in determining calcium, since only between 4 and 5 mg of this element is present in the extract.

As for iron and copper in the extracts, the amounts found were so small that the results were not sufficiently reliable to include in detail. There was a difference, however, in the amounts of these constituents leached by the two cooking methods. The amount of iron leached during boiling varied from 0.8 to 1.7 mg, whereas, that leached during steaming varied between 0.16 and 0.50 mg. The amount of copper leached during boiling was from 0.4 to 0.7 mg; that leached during steaming varied between 0.07 and 0.20 mg. Leaching of these elements was higher early in the storage period.

*Effect of Storage on the Nitrogen Content of Peas.*—The dried, cooked, and uncooked peas were analyzed for total nitrogen, with results shown in table 7. The nitrogen content of the uncooked peas did not vary appreciably throughout the storage period. Except for the master sample,

which constitutes the initial point, the curve for the uncooked peas would form practically a straight line. More nitrogen was found in the uncooked peas than in the cooked, and more in the steamed peas than in the boiled. Obviously, more nitrogenous material was leached out during the boiling than during the steaming; and more with either method of cooking than with asparagus.

Tests were made for nitrate in the master sample, but none was found.

A test was also made to determine whether or not nitrogen is lost during the drying of peas. Some peas were bought from the market and

TABLE 8

CRUDE-FIBER CONTENT AND PUNCTURE TEST ON UNCOOKED PEAS

(Storage temperature 34° F)

| Storage period | Crude-fiber content per 400 grams of peas | | Puncture tests,* mean | Storage period | Crude-fiber content per 400 grams of peas | | Puncture tests,* mean |
|---|---|---|---|---|---|---|---|
| | Mean | s† | | | Mean | s† | |
| *hours* | *grams* | *grams* | *units‡* | *hours* | *grams* | *grams* | *units‡* |
| 0 | 6.28 | 0.50 | 4.94 | 96 | 7.29 | 0.12 | 11.08 |
| 24 | 7.18 | 0.23 | 7.08 | 168 | 7.63 | 0.29 | 12.10 |
| 48 | 7.29 | 0.26 | 7.62 | 240 | 7.40 | 0.06 | 13.94 |

* $F$=25.53. One per cent point 3.08. Minimum significant difference 1.94 puncture units.
† $s$=Estimated standard deviation.
‡ See page 310 of text.

cooked. The cooked peas were placed in a large flask and heated over a steam bath. At the same time, air, which had previously been bubbled through concentrated sulfuric acid, was passed through the flask and bubbled again through concentrated sulfuric acid. When the peas had dried for 2 days, this acid solution was then analyzed for ammoniacal nitrogen, and less than 1 mg was found. Since this is nearly within the limit of accuracy of detecting nitrogen, one may safely conclude that little or no nitrogen is lost during drying—especially if the peas are dried at a low temperature, so that no scorching takes place.

With peas more nitrogenous material was leached during either method of cooking than with asparagus.

*Effect of Storage on Crude-Fiber Content of Peas.*—The crude-fiber content per 400 grams of peas and the mean puncture units for uncooked peas at each storage period are shown in table 8. The crude-fiber content increased with storage, the most rapid increase occurring during the first 24 hours of storage. The difference in mean puncture units between peas fresh from the field (0 hours, 4.94 units) and those stored 24 hours (7.08 units) is 2.14; and between 0 hours and 240 hours, the difference is 9.00 units; both of these differences are statistically significant. Be-

tween 96 and 168 hours, on the other hand, the difference is only 1.02 units and is not considered statistically significant. The calculated differences show that peas grow measurably firmer with age and that differences in twelve out of fifteen cases (that is, all the possible combinations that can be made between the six means given in table 8) are statistically significant.

## SUMMARY AND CONCLUSIONS

Data presented show the effect of storage at 35° F for various periods of time on the leaching of magnesium, calcium, phosphorus, and nitrogen from asparagus (Palmetto variety) and peas (Giant Stride variety) when boiled and steamed, and on the crude-fiber content.

At each storage period greater amounts of total solids were leached from asparagus and peas during boiling than during steaming. Approximately four times as much solid material—which of course includes sugars, starches, proteinaceous material, as well as salts—was leached from boiled asparagus as from steamed, and approximately three times as much from boiled peas as from steamed. These results are in accord with those of Peterson and Hoppert (*3*), who found that the mineral losses increased when vegetables were boiled; and with those of Talenti and Ragno (*5*), who concluded that steaming is superior to boiling in water because it extracts one-half less organic matter and two-thirds less mineral matter.

With asparagus, approximately four times as much magnesium, calcium, and phosphorus were leached by boiling as by steaming. With peas, approximately twice as much magnesium and phosphorus were leached during boiling as during steaming, but only slightly more calcium.

With asparagus, the order of decreasing percentage leached, both for boiling and for steaming, was magnesium, calcium, and phosphorus; with peas, however, the order was magnesium, phosphorus, and calcium.

The rate of leaching from asparagus during both processes of cooking decreased rapidly during the first 48 hours and then gradually decreased with increase of storage period. With the peas, the rate of leaching gradually decreased throughout the storage period; there was no marked initial decrease. The initial decrease in the case of the asparagus probably results from a rapid transference of the elements into less-soluble organic forms.

More nitrogen was leached from boiled peas and asparagus than from steamed. More nitrogenous material was leached from peas during either method of cooking than from asparagus.

The crude-fiber content of uncooked asparagus at the various storage

periods did not vary significantly. Peas, however, grow measurably firmer with storage. The most rapid increase in crude-fiber content occurred during the first 24 hours.

Peas and asparagus harvested and stored under suitable temperature and humidity conditions for more than 48 hours, retain more of their mineral constituents when cooked either by boiling or steaming than when cooked immediately after harvesting. This is an extremely important point; for peas and asparagus, whether marketed fresh, canned, or quick-frozen, are usually held over several hours or even a few days before being processed.

The rate of leaching is dependent not only on the particular element but also on the amount of water coming in contact with and passing through the vegetable. For this reason, steamed peas and asparagus retain more of their constituent elements than boiled.

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# BUFFERING ACTION OF NONACID VEGETABLES

GEORGE L. MARSH

# BUFFERING ACTION OF NONACID VEGETABLES[1]

GEORGE L. MARSH[2]

JUICES FROM ALL PLANT TISSUES exhibit more or less ability to resist changes in pH value on the addition of strong acids or bases (*15*).[3] The substances responsible for this buffering capacity have not been completely defined largely owing to the complexity of the systems involved. The complex nature of the extracted juices increases the difficulties of identifying the specific buffer substances. In those plant juices which have been investigated most extensively, the buffer capacity has been ascribed to organic acids (*10*), dialyzable acid-salt systems (*14*), proteins (*6*), and acid phosphates (*9*). Probably the buffering is also due to the adsorption reactions of the colloids as well as to dissociation of the weak acids or bases that may be present.

The buffering capacity of nonacid vegetables is of particular importance in connection with the use of acidified brines in canning. Cruess, Fong, and Liu (*4*) found that nonacid vegetables canned with a sufficiently acid brine to bring their pH value below 4.5 may be readily pasteurized in boiling water and need not be processed in steam-heated pressure cookers. These investigators found, however, that the nonacid vegetables exerted a pronounced buffering action when canned in brines acidified with hydrochloric, citric, or acetic acids: a marked rise in pH value of the added brine occurred during heating. The increase was much greater than could be accounted for by diffusion, and they concluded that the rise in pH value was probably caused principally by the action of buffer substances dissolved from the vegetables. Bigelow and Cathcart (*2*) had also noted that when beans were canned with tomato sauce there was an increase in pH value of the sauce which they stated to be "due to diffusion of acids into the beans."

Aside from the work of the above investigators, no detailed study of the buffer capacity of nonacid vegetables is available.

For the past several seasons, some attention has been devoted to a study of this problem. Experiments were undertaken to determine the effect of heat on the buffering capacity of the juices extracted from several vegetables, to determine the buffer index of the vegetable juices, and in addition to investigate in greater detail the changes in pH value during

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[3] Italic numbers in parentheses refer to "Literature Cited," at the end of this paper.

the canning of vegetables in brines acidified to varying degrees with citric, acetic, and hydrochloric acids. The results of these various experiments are reported in this paper.

## pH VALUES OF EXPRESSED JUICES

*Methods of Determining pH Values.*—In the experiment to determine the effect of heat on the buffer capacity of nonacid vegetable juices, green-asparagus, pea, and string-bean juices were extracted from the fresh vegetables of midseason maturity during 1932, by coarsely grinding and pressing. The juice was preserved in the frozen state in cans at a storage temperature of —17° C. Before use, the juices were thawed quickly in the cans in warm water and brought to room temperature. Several 100-cc portions of each vegetable juice were measured into 100-cc volumetric flasks graduated at 100 and 110 cc. Amounts of 0.1*N* or *N* HCl and citric acids corresponding to those shown in tables 1 and 2 respectively and amounts of 0.1*N* or *N* NaOH corresponding to those shown in table 3 were added to the samples. Each sample was then diluted to 110 cc, the contents thoroughly mixed, and the pH value determined on duplicate portions of each sample. The remainder of each sample was heated at 100° C for 1 hour in tightly sealed, 120-cc glass jars. After cooling to room temperature, the pH value of duplicate portions of the thoroughly mixed contents of each jar was again determined.

The pH measurements were made with a Hildebrand-type hydrogen electrode at a temperature of approximately 25° C. The electrodes were frequently replatinized and checked against standard buffer solutions. After each measurement in the alkaline range, the electrode was removed and dipped in dilute hydrochloric acid and thoroughly washed with water before proceeding with the next sample. The electrode became poisoned less rapidly when this procedure was followed. Commercial compressed hydrogen, purified by being passed through an absorption train of alkaline pyrogallol and soda lime, was used. The results obtained are probably accurate to 0.02 pH unit.

*pH Values after Heating with Acid or Alkali.*—The data obtained are presented in tables 1, 2, and 3. The curves in figure 1 are plotted from the data obtained with pea juice. Heating in the presence of acid lowered the pH value of green-asparagus and string-bean juices acidified with small amounts of hydrochloric acid but raised the pH values of these juices when sufficient acid was added before heating to bring the pH of green-asparagus juice below 3.0 and of string-bean juice below 4.0. Heating raised the pH value of pea juice acidified with hydrochloric acid. The pH changes that occurred on heating pea and green-asparagus juices

acidified with citric acid were inconsistent and slight and did not exceed 0.2 pH unit. On the other hand, a marked lowering of the pH occurred on heating juices containing added sodium hydroxide when the initial pH value exceeded 6.5.

Vegetable juices therefore behave like other biological fluids with respect to change in pH value when heated in the presence of acid or alkali.

TABLE 1

EFFECT OF HEAT ON THE pH VALUE OF GREEN-ASPARAGUS, PEA, AND STRING-BEAN JUICES ACIDIFIED WITH VARIOUS AMOUNTS OF HCl

| *N* HCl added | Green asparagus | | Pea | | String bean | |
|---|---|---|---|---|---|---|
| | Before heating | After heating | Before heating | After heating | Before heating | After heating |
| *cc* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| 0.00 | 6.24 | 6.04 | 5.40 | 5.34 | 5.98 | 5.74 |
| 0.10 | 6.06 | 5.93 | 5.29 | 5.31 | 5.88 | 5.68 |
| 0.20 | 5.88 | 5.74 | .... | .... | 5.78 | 5.55 |
| 0.30 | 5.69 | 5.62 | 5.20 | 5.25 | 5.66 | 5.34 |
| 0.40 | 5.48 | 5.47 | .... | .... | 5.57 | 5.38 |
| 0.50 | 5.31 | 5.25 | 5.14 | 5.13 | 5.49 | 5.29 |
| 0.75 | 5.00 | 4.88 | .... | .... | 5.33 | 5.11 |
| 1.00 | 4.66 | 4.47 | 4.92 | 4.99 | 5.13 | 4.96 |
| 2.00 | 3.96 | 3.89 | 4.65 | 4.65 | 4.66 | 4.57 |
| 3.00 | .... | .... | 4.43 | 4.46 | 4.26 | 4.32 |
| 4.00 | 2.96 | 3.14 | 4.19 | 4.25 | .... | .... |
| 5.00 | .... | .... | 3.95 | 4.04 | 3.63 | 3.68 |
| 7.50 | .... | .... | 3.51 | 3.61 | 3.00 | 3.07 |
| 10.00 | .... | .... | 3.09 | 3.22 | 2.43 | 2.45 |

TABLE 2

EFFECT OF HEAT ON THE pH VALUE OF GREEN-ASPARAGUS AND PEA JUICES ACIDIFIED WITH VARIOUS AMOUNTS OF CITRIC ACID

| *N* citric acid added | Green asparagus | | Pea | |
|---|---|---|---|---|
| | Before heating | After heating | Before heating | After heating |
| *cc* | *pH* | *pH* | *pH* | *pH* |
| 0.00 | 6.24 | 6.04 | 5.40 | 5.35 |
| 0.10 | 6.06 | 5.89 | .... | .... |
| 0.20 | .... | .... | 5.27 | 5.29 |
| 0.30 | .... | .... | 5.23 | 5.24 |
| 0.40 | .... | .... | 5.17 | 5.19 |
| 0.50 | 5.45 | 5.40 | 5.14 | 5.17 |
| 0.75 | .... | .... | 5.08 | 5.10 |
| 1.00 | 4.97 | 4.93 | 5.00 | 5.03 |
| 2.00 | 4.53 | 4.47 | 4.78 | 4.75 |
| 3.00 | 4.26 | 4.20 | 4.62 | 4.57 |
| 4.00 | 4.05 | 4.02 | 4.50 | 4.42 |
| 5.00 | 3.96 | 4.00 | 4.39 | 4.38 |
| 7.50 | 3.68 | 3.71 | 4.17 | 4.17 |
| 10.00 | 3.48 | 3.50 | 4.02 | 3.95 |

TABLE 3

Effect of Heat on the pH Value of Green-Asparagus, Pea, and String-Bean Juices Alkalized with Various Amounts of NaOH

| *N* NaOH added | Green asparagus | | Pea | | String bean | |
|---|---|---|---|---|---|---|
| | Before heating | After heating | Before heating | After heating | Before heating | After heating |
| *cc* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| 0.00 | 6.24 | 6.04 | 5.40 | 5.34 | 5.98 | 5.74 |
| 0.10 | 6.37 | 6.21 | 5.46 | 5.44 | 6.11 | 5 95 |
| 0.20 | .... | .... | 5.50 | 5.49 | 6.22 | 6.04 |
| 0.30 | 6.68 | 6.50 | 5.58 | 5.54 | 6.33 | 6.12 |
| 0.40 | .... | .... | 5.63 | 5.57 | 6.45 | 6.23 |
| 0.50 | 6.91 | 6.76 | 5.69 | 5.59 | 6.57 | 6.36 |
| 0.75 | .... | .... | 5.80 | 5.70 | 6.84 | 6.62 |
| 1.00 | 7.43 | 7.06 | 6.03 | 5.91 | 7.14 | 6.90 |
| 2.00 | 8.33 | 7.61 | 6.70 | 6.29 | 8.15 | 7.62 |
| 3.00 | 8.80 | 7.80 | 7.37 | 6.67 | 8.75 | 7.90 |
| 4.00 | .... | .... | 7.93 | 7.13 | 9.18 | 8.36 |
| 5.00 | .... | .... | 8.21 | 7.41 | 9.59 | 9.00 |
| 7.50 | .... | .... | 8.53 | 7.92 | .. | |
| 10.00 | .... | .... | 8.68 | 8.30 | 10.94 | 10.50 |

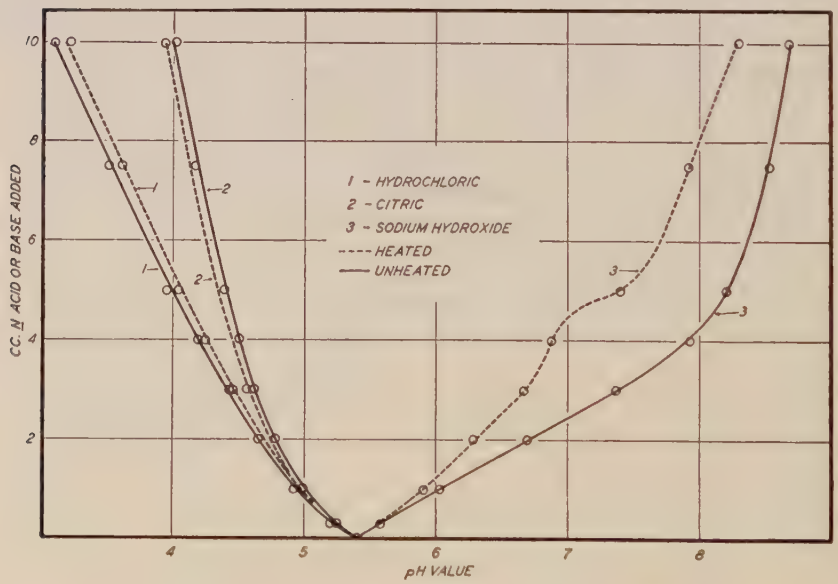


Fig. 1.—The effect of heat upon the pH value of pea juice after the addition of acid or base.

It is well known that prepared bacteriological media change in pH on heating, particularly when their initial pH values are above 7.0 (*1*). Cruess, Richert, and Irish (*5*) give data showing the changes in pH value upon sterilization of media prepared from bacto-peptone, Libby's beef extract, glucose, and small amounts of inorganic salts, and adjusted to various pH values with citric and sodium hydroxide. Below pH 4.0, the media, on heating, increases slightly in pH, while above pH 4.0, it de-

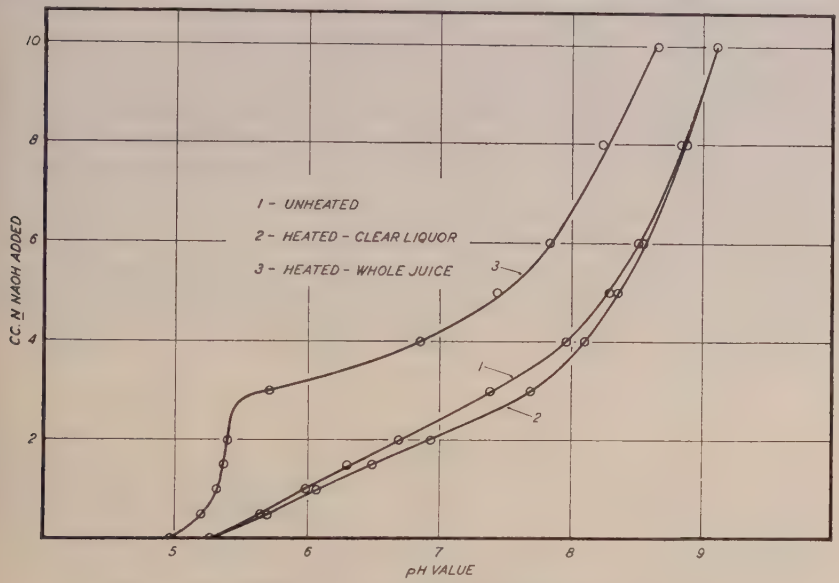


Fig. 2.—The effect of heat upon the potentiometric titration of different fractions of pea juice with NaOH.

creases, the decrease becoming more marked the higher the original pH value. McClung (*11*) has also called attention to the occurrence of these phenomena in certain vegetable-extract bacteriological media. The change in pH value occurring upon heating in biological fluids adjusted to high pH values is probably largely due to the formation of acidic materials by the decomposition of sugars by the alkali—a change known to occur in pure sugar solutions at high pH values (*3, 7, 12*).

*pH Values of Supernatant Liquor of Pea Juice.*—During the heating of the samples, certain substances contained in the raw juices coagulated and precipitated from solution. The supernatant liquor was straw-colored and brilliantly clear. The buffer capacity of pea juice free from its heat-coagulable colloidal material was tested in comparison with the unheated juice and with the entire, well-mixed, heated juice containing the resuspended coagulum.

A 500-cc portion of the previously prepared pea juice was placed in a tightly sealed glass jar, heated for 1 hour at 100° C, and cooled to room temperature. The heat-coagulated material was resuspended by shaking and stirring and two 100-cc aliquots were withdrawn. The remainder was filtered. Alkaline potentiometric titrations were then conducted on duplicate portions of each of the above samples and on duplicate portions of the unheated raw juice. The pH values were recorded after each addition of *N* NaOH measured from a 10-cc microburette. Great difficulty was encountered in the determination of the titration curve of the heated sample containing the resuspended coagulum, because the electrode poisoned rapidly. Therefore, a freshly platinized electrode was used to determine the pH value after each addition of *N* NaOH.

The average values of the results obtained are given in figure 2. Heat caused little change in the initial pH value of the clear liquor, whereas it appreciably lowered the initial pH value of the whole juice. The clear liquid shows less buffer action toward added NaOH than does the unheated juice. The heated whole juice shows a very strong buffering action in the range pH 5.0 to 5.7; markedly more than that of the unheated juice.

## BUFFER INDEX OF EXPRESSED NONACID VEGETABLE JUICES

*Methods of Calculating Index and of Determining pH Values.*—Van Slyke (*16*) has established the buffer index as a measure expressing buffer capacities of various solutions. The unit adopted by Van Slyke is the differential ratio $\frac{dB}{d\text{pH}}$, or $\beta$, which is defined as the rate of change in amount of strong base added with change in pH value produced. If an acid is added, the values $d$B and $d$pH are negative; hence the ratio, the Van Slyke $\beta$, always has a positive value. This differential ratio is usually difficult to measure, and the ratio $\frac{\Delta B}{\Delta \text{pH}}$, or B, where each of these values is a measurable increment, is commonly used. This differs from $\beta$ in being the slope of the intercept between any two points of a B-versus-pH graph instead of the slope of the tangent to the curve at one pH value. The smaller the pH interval used in calculating B, the closer will it approach $\beta$ in value. In the results reported herein, the data obtained were plotted on large-scale graphs and for each $\Delta\text{pH} = 0.2$, the corresponding $\Delta$B was measured, and plotted against the average pH.

The juices used for the determination of the buffer index were prepared from vegetables of midseason maturity during 1933 by coarsely grinding and pressing in an American Utensil expeller press. Finely

TABLE 4

CHANGES IN pH VALUE OF VEGETABLE JUICES ON ADDITION OF HCl

| HCl per liter | String-bean juice | Green-asparagus juice | Spinach juice | Pea juice | White-asparagus juice | Artichoke juice |
|---|---|---|---|---|---|---|
| *mols* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| 0.000 | 5.92 | 5.40 | 6.56 | 5.37 | 6.20 | 5.58 |
| 0.001 | 5.70 | 5.37 | 6.46 | 5.30 | 5.83 | 5.56 |
| 0.002 | .... | 5.28 | 6.40 | 5.23 | 5.68 | 5.47 |
| 0.003 | 5.49 | 5.19 | 6.32 | 5.22 | 5.45 | 5.40 |
| 0.004 | .... | 5.10 | 6.24 | 5.18 | 5.41 | 5.32 |
| 0.005 | .... | 5.05 | 6.15 | 5.13 | 5.29 | 5.25 |
| 0.006 | 5.24 | 4.96 | 6.06 | 5.10 | 5.17 | 5.19 |
| 0.008 | .... | 4.81 | 5.87 | 5.01 | 4.97 | 5.05 |
| 0.010 | 5.01 | 4.70 | 5.65 | 4.94 | 4.79 | 4.97 |
| 0.015 | 4.76 | 4.43 | 5.21 | 4.77 | 4.49 | 4.68 |
| 0.020 | 4.60* | 4.22 | 4.86 | 4.63 | 4.16 | 4.53 |
| 0.030 | 4.19 | 3.87 | 4.38 | 4.37 | 3.65 | 4.18 |
| 0.040 | 3.85 | 3.56 | 4.05 | 4.15 | 3.20 | 3.87 |
| 0.050 | .... | 3.28 | 3.77 | 3.95 | 3.87 | 3.57 |
| 0.060 | 3.23 | 3.01 | 3.53 | 3.74 | 2.59 | 3.27 |
| 0.080 | 2.75 | 2.59 | 3.11 | 3.36 | 2.12 | 2.80 |
| 0.100 | 2.31 | 2.25 | 2.68 | 3.04 | 1.81 | 2.41 |

* 0.021 mols.

TABLE 5

CHANGES IN pH VALUE OF VEGETABLE JUICES ON ADDITION OF NaOH

| NaOH per liter | String-bean juice | Green-asparagus juice | Spinach juice | Pea juice | White-asparagus juice | Artichoke juice |
|---|---|---|---|---|---|---|
| *mols* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| 0.000 | 5.92 | 5.40 | 6.56 | 5.37 | 6.20 | 5.58 |
| 0.001 | 6.05 | 5.42 | 6.74 | 5.41 | 6.42 | 5.59 |
| 0.002 | 6.16 | 5.47 | 6.84 | 5.47 | 6.59 | 5.65 |
| 0.003 | 6.27 | 5.53 | 6.95 | 5.53 | 6.72 | 5.70 |
| 0.004 | 6.39 | 5.58 | 7.09 | 5.58 | 6.89 | 5.76 |
| 0.005 | 6.50 | 5.65 | 7.21 | 5.65 | 7.03 | 5.82 |
| 0.006 | 6.62 | 5.71 | 7.34 | 5.71 | 7.17 | 5.86 |
| 0.008 | 6.84 | 5.86 | 7.61 | 5.86 | 7.49 | 5.97 |
| 0.010 | 7.08 | 5.99 | 7.88 | 5.99 | 7.77 | 6.08 |
| 0.015 | 7.60 | 6.30 | 8.45 | 6.30 | 8.23 | 6.37 |
| 0.020 | 8.07 | 6.63 | 8.84 | 6.68 | 8.53 | 6.56 |
| 0.030 | 8.61 | 7.38 | .... | 7.41 | 8.97 | 7.21 |
| 0.040 | 9.04 | 7.89 | .... | 7.97 | 9.34 | 7.52 |
| 0.050 | 9.43 | 8.26 | .... | 8.29 | 9.72 | 7.82 |
| 0.060 | 9.80 | 8.50 | .... | 8.51 | 10.13 | 8.04 |
| 0.080 | 10.51 | 8.79 | .... | 8.84 | 10.72 | 8.46 |
| 0.100 | 10.96 | 9.02 | .... | 9.11 | 10.97 | 8.87 |

ground suspended material was separated by sieving through several layers of cheesecloth. They were preserved by freezing until used. The frozen samples were thawed, and duplicate 100-cc portions of each vegetable juice were titrated with *N* HCl and *N* NaOH added from a 10-cc microburette. The pH values after each addition of acid or base were determined by means of a hydrogen electrode.

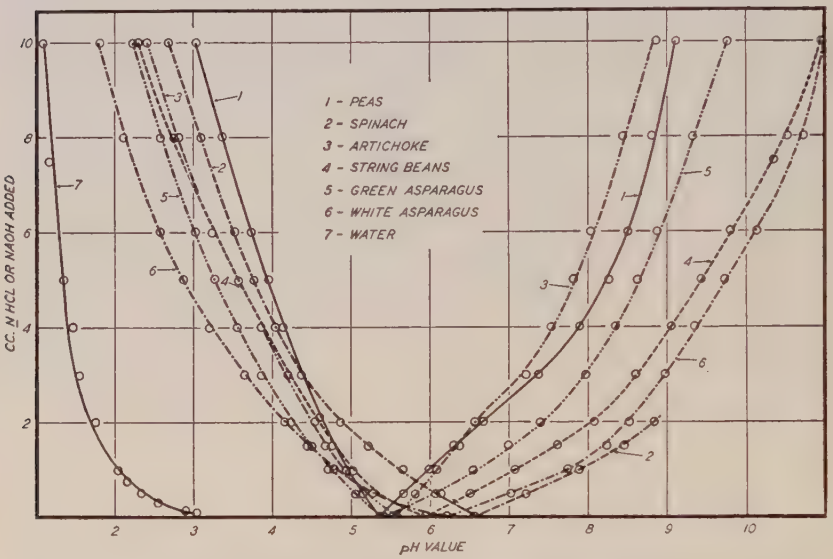


Fig. 3.—The potentiometric titration curves of 100-cc portions of vegetable juices with HCl and NaOH.

*Comparison of Buffer-Index Curves.*—The results of the potentiometric titrations on the expressed juices of the various vegetables are recorded in tables 4 and 5 and shown graphically in figure 3. The vegetables used differ markedly in their buffer capacity. The juices of peas and of artichokes resist change in pH upon addition of acid or base to far greater extent than do those of white asparagus and spinach.

The Van Slyke B values plotted against pH are shown in figure 4. These curves indicate that there are present in the various vegetable juices certain substances that exert a definite buffering action in the pH range 3.5 to 4.2, also in the range from pH 6.0 to 7.5, and again in the range from pH 8.5 to 10.0. The type of curve obtained would indicate that the buffering action is not due to a simple system but is in all probability due to a very complex mixture of buffer substances.

Oakley and Krantz (*13*) report for tomato juice at pH 3.5 a B value of 0.047 and a $\beta$ value of 0.033. This latter value, however, is in error and

when calculated from the data they present should be no lower than 0.045. Their curve for tomato juice also shows the presence of definite buffer substances in the range of pH from 3.5 to 4.0. Pea juice, therefore, very closely approaches tomato juice in its buffer capacity toward added strong acid.

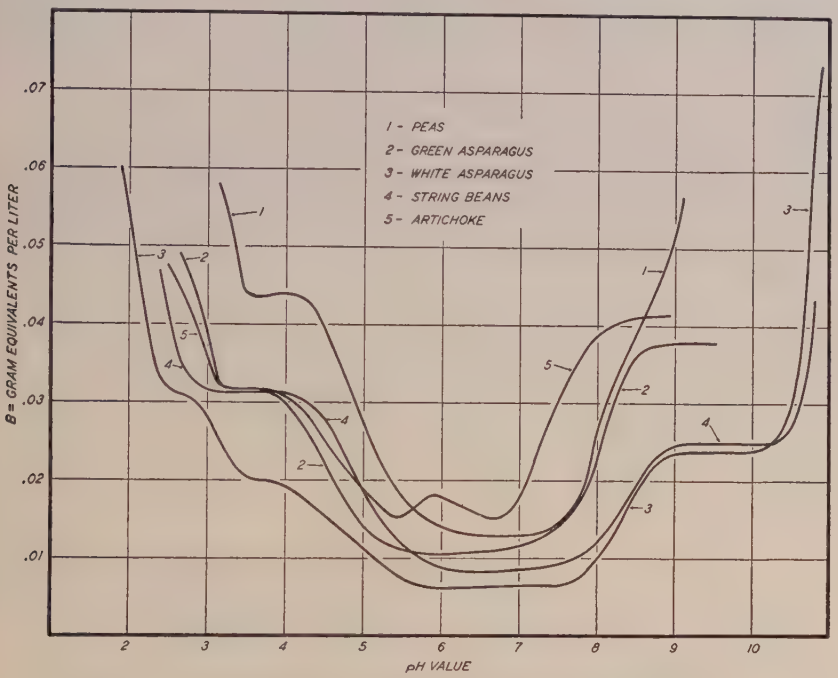


Fig. 4.—The buffer index of vegetable juices at various pH values.

## pH VALUES AND TOTAL ACIDITY OF VEGETABLES CANNED IN ACIDIFIED BRINES

*Review of Previous Work.*—Cruess, Fong, and Liu (*4*) and many others have shown that the death temperature and death time of heat-resistant spore-bearing bacteria are greatly affected by the pH of the medium. Their results indicate that if it is feasible to reduce the pH value of the vegetable below 4.5 by the addition of acid to the brine, the present industrial method of sterilizing vegetables of low hydrogen-ion concentration by steam under pressure—which results in injury to texture, flavor, and color—need not be used.

As was pointed out by these investigators and confirmed by the results already presented, however, nonacid vegetables exert pronounced buffering action toward added acids. During processing, a marked increase

in pH value of the added acidified brine occurs as a result of the intimate mixing of the juice of the vegetable and the acidified brine. Mixing is made possible by the heat applied during processing, which kills the living cellular tissue and thereby causes it to lose its selective permeability. Since the pH value after heating was found by the above investigators to be more significant in relation to the effect on the heat resistance of spores than was the initial pH value of the brine and since they did not make a direct study of the changes occurring in pH value of acidified brines during the canning procedure, it was thought that a detailed study of the factors responsible for the observed changes was desirable. The following data were obtained from samples packed experimentally during three different seasons. Each season the vegetables used for the tests were of approximately the same maturity as those used the preceding season but were grown under different climatic and soil conditions.

*Methods of Canning and Acidifying.*—The vegetables were prepared for canning in the usual manner, and in the tests made during the first two seasons approximately 150-gram portions were placed in 8-ounce cans. There were then added approximately 100-cc portions of brine containing 2 per cent of NaCl and acidified with various amounts of acid. After the addition of the acidified brine, the cans were heated for 3 minutes in steam at 100° C, sealed, and processed for 1 hour in water at 100° C.

During the third season, the samples were prepared in a more accurate manner for the purpose of attempting to establish an acid balance in the samples before and after canning. Therefore an approximately constant amount of vegetable was placed in previously tared 8-ounce cans, the samples were weighed, brine was then added, and the samples reweighed. The lids were crimped in place by a light first roll, the cans were then exhausted 10 minutes at 100° C, given a second roll to seal them airtight, and pasteurized for 30 minutes at 100° C. After canning, all samples were cooled quickly in running water and were subsequently stored at 0° C until removed for pH determinations. All samples were packed in triplicate, two samples for pH determinations and the third for visual examination and organoleptic tests.

Samples the first season were prepared from green asparagus, string beans, and peas packed with brines containing 0.00, 0.10, 0.20, 0.30, 0.40, 0.50, 0.75, 1.00 and 2.00 grams of citric acid per 100 cc. The following season, green asparagus, string beans, peas, spinach, and a mixed-vegetable sample were packed with brine prepared by adding 0.0, 2.5, 5.0, 7.5, 10.0 and 15.0 grams of citric acid and 25, 50, 75, 100, 125, 150 and 200 cc of 40-grain vinegar to 1,000-cc portions of 2 per cent NaCl brine. The

mixed-vegetable pack, referred to as "vegetable salad," was composed of peas, string beans, green asparagus, and carrots in equal proportion by weight, with a small piece of pimiento in each can. The actual acid concentration of the various brines are shown in table 7. Only pea and string-bean samples were prepared for the tests made during the third season. These vegetables were canned in brines prepared by dissolving 20 grams of NaCl and the required amount of acid in a small volume of water, transferring to 1,000-cc volumetric flasks, and making up to volume. The total acid concentration and the pH value of the brines prepared for this test are given in table 8, page 331.

In addition, a large quantity of the peas and string beans used during the third season were coarsely ground, frozen and thawed, and the juice extracted by pressure. The juice obtained was preserved in freezing storage at —17° C.

Preliminary tests showed that after canning and storage the pH values of the brine and that of the juice expressed from the vegetables were practically identical. On this account, only the pH values of the brines drained from the vegetables were determined in most cases. The following values indicate the agreement in pH values of the juice expressed from the vegetables and that of the drained brines.

| | Drained brine, pH | Expressed juice, pH |
|---|---|---|
| Peas | 5.96 | 6.00 |
| String beans | 5.90 | 5.98 |
| Asparagus | 5.51 | 5.54 |

The pH determinations on the first two seasons' samples were made with the hydrogen electrode, but those on the last set were made with the quinhydrone electrode. Tests indicated that the latter gave results that were just as reliable. It has the advantages that equilibrium is more quickly attained and that, unlike the hydrogen electrode, it is not easily poisoned by the solutions used in these studies.

*Amount of Change in pH after Processing, First Two Seasons' Tests.*— The results of the pH determinations on the first two season's samples are reported in tables 6 and 7, and part of the data of table 7 is graphically presented in figures 5 and 6.

An examination of table 6 indicates that during the canning and processing of vegetables in acidified brines, there occurs a decrease in pH value of the vegetable tissue, which is directly related to the total amount of acid added; and an increase in pH value of the brine, which is related to the initial pH value of the brine—the nature of the vegetable under examination determining the magnitude of the changes. Peas exert a

TABLE 6

Change of pH Value of Asparagus, String Beans, and Peas Canned in Brines Acidified with Citric Acid

| Total acid of brines before canning | pH value of added brine | Green asparagus (initial pH=5.60) | | | String beans (initial pH=5.90) | | | Peas (initial pH=6.40) | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | pH after processing | Change in pH of vegetable | Change in pH of brine | pH after processing | Change in pH of vegetable | Change in pH of brine | pH after processing | Change in pH of vegetable | Change in pH of brine |
| *grams per 100 cc* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| 0.00 | .... | 5.45 | 0.15 | .... | 5.57 | 0.33 | .... | 6.38 | 0.02 | |
| 0.10 | 2.70 | 5.10 | 0.50 | 2.40 | 5.18 | 0.72 | 2.48 | 5.96 | 0.44 | 3.26 |
| 0.20 | 2.44 | 4.79 | 0.81 | 2.35 | 4.80 | 1.10 | 2.36 | 5.48 | 0.92 | 3.04 |
| 0.30 | 2.36 | 4.58 | 1.02 | 2.22 | 4.65 | 1.25 | 2.29 | 5.30 | 1.10 | 2.94 |
| 0.40 | 2.28 | 4.47 | 1.13 | 2.19 | 4.48 | 1.42 | 2.20 | 5.06 | 1.34 | 2.78 |
| 0.50 | 2.23 | 4.23 | 1.37 | 2.00 | 4.19 | 1.71 | 1.96 | 4.77 | 1.63 | 2.54 |
| 0.75 | 2.12 | 4.07 | 1.53 | 1.95 | 4.09 | 1.81 | 1.97 | 4.57 | 1.83 | 2.45 |
| 1.00 | 2.05 | 3.98 | 1.62 | 1.93 | 3.83 | 2.07 | 1.78 | 4.36 | 2.04 | 2.31 |
| 2.00 | 1.87 | 3.23 | 2.37 | 1.36 | 3.27 | 2.63 | 1.40 | 3.72 | 2.68 | 1.85 |

TABLE 7

Changes in pH Value of Vegetables Canned in Brines Acidified with Citric Acid and in Those Acidified with Vinegar (Acetic Acid)

| Acid added to 1,000 cc of brine | Total acid of brines before canning | pH value of added brine | pH values after processing* | | | | |
|---|---|---|---|---|---|---|---|
| | | | Peas | Green asparagus | String beans | Vegetable salad | Spinach |
| | *grams per 100 cc* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| No added acid | .... | .... | 6.35 | 5.61 | 5.49 | 5.56 | 5.73 |
| 2.5 grams citric acid | 0.22 | 2.54 | 5.55 | 4.56 | 4.71 | 4.76 | 5.32 |
| 5.0 grams citric acid | 0.44 | 2.31 | 5.02 | 4.31 | 4.24 | 4.35 | 5.05 |
| 7.5 grams citric acid | 0.68 | 2.17 | 4.69 | 3.95 | 4.00 | 4.10 | 4.80 |
| 10 grams citric acid | 0.89 | 2.12 | 4.50 | 3.75 | 3.81 | 3.86 | 4.64 |
| 15 grams citric acid | 1.34 | 2.03 | 4.19 | 3.35 | 3.49 | 3.62 | 4.40 |
| 25 cc vinegar | 0.24 | 2.99 | 5.28 | 4.65 | 4.64 | 4.68 | |
| 50 cc vinegar | 0.48 | 2.87 | 4.96 | 4.39 | 4.32 | 4.53 | |
| 75 cc vinegar | 0.70 | 2.80 | 4.84 | 4.25 | 4.18 | 4.36 | |
| 100 cc vinegar | 0.90 | 2.73 | 4.65 | 4.14 | 4.12 | 4.32 | |
| 125 cc vinegar | 1.10 | 2.70 | 4.57 | 4.03 | 4.02 | 4.27 | |
| 150 cc vinegar | 1.32 | 2.66 | 4.44 | 4.00 | 3.98 | 4.13 | |
| 200 cc vinegar | 1.66 | 2.61 | 4.28 | 3.92 | 3.86 | 4.08 | |

* Average of the determinations on duplicate samples.

greater buffer effect than string beans or green asparagus, which two vegetables show nearly identical buffering power.

The pH value of the string beans and green asparagus canned in non-acidified brine likewise decreased in pH value during processing, but very little change occurred in the peas. This observation is in accordance with the findings of Bigelow and Catheart, in which all vegetables studied by them—peas included, however—decreased in pH value dur-

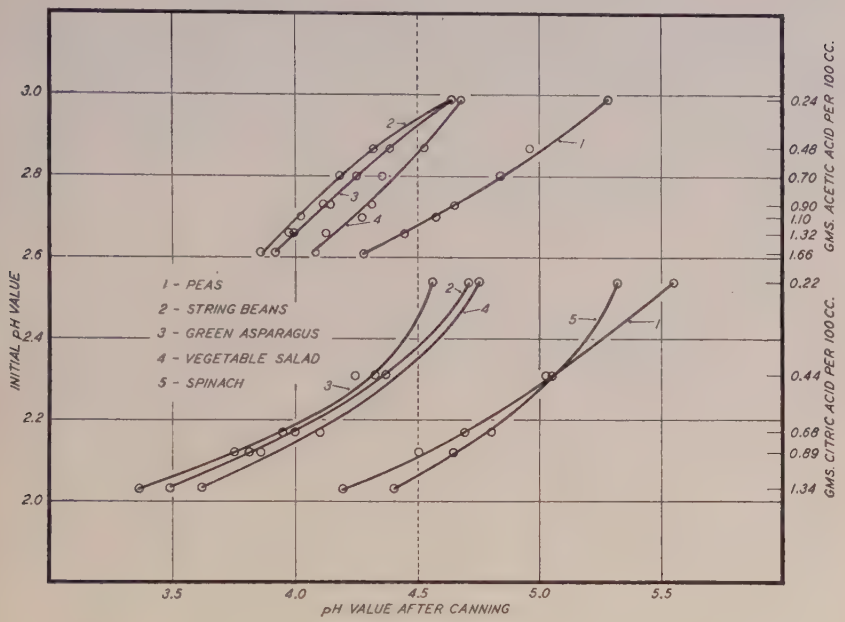


Fig. 5.—Changes in pH value occurring in the acidified brines during processing at 100° C.

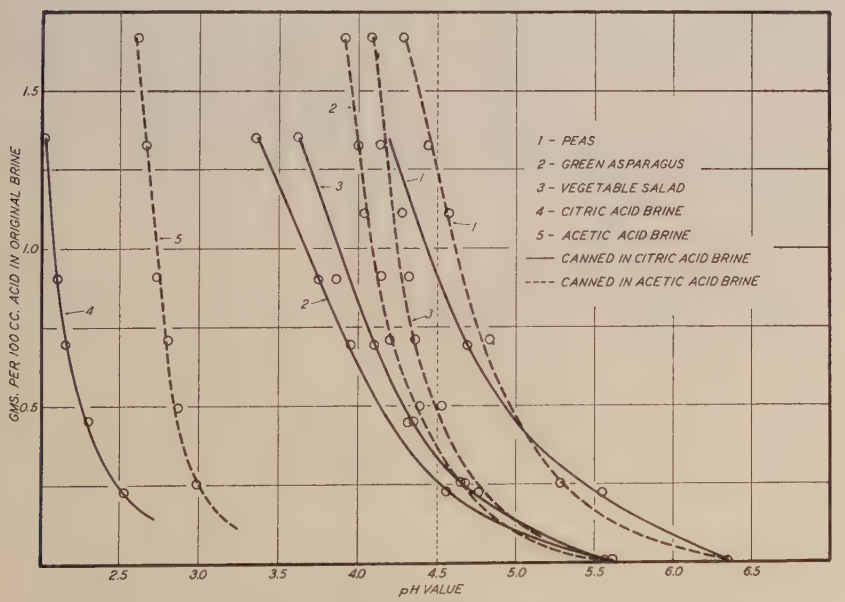


Fig. 6.—The pH value of vegetables canned in various concentrations of acidified brine after processing at 100° C.

ing processing; they believed this to be due to the formation of acidic substances such as carbon dioxide or hydrogen sulfide, to a reaction between the proteins and aldehydes rendering the former more acidic, or to the precipitation of buffer substances by heat. Data will be presented to show that acidic constituents are formed during heating of string-bean juice but not during the heating of pea juice.

As shown in figure 5, acetic acid brines of low initial total acid concentration produce slightly lower pH values after processing than citric acid brines of the same total acid concentration but of lower pH value. This observed difference in behavior, in which the weaker acid causes a greater change in pH value of the product, is in this instance probably due to the buffering effect of compounds other than acetic acid contained in the 40-grain cider vinegar used as a source of acetic acid. Other tests in which acetic acid brines were prepared from glacial acetic acid show that citric and acetic acid brines of similar low total acid concentration affect the pH value of the vegetable tested to the same extent. At higher total acid concentrations, however, the decrease in pH value is greatest in the vegetables canned in citric acid brines.

On the other hand, owing to the differences in degree of ionization, acetic acid brines lower the pH value of the vegetable to a greater extent than citric or hydrochloric acid brine of the same initial pH value; and citric acid in turn decreases the pH value to a greater extent than hydrochloric. Since at any pH value in which a comparison is made of the three acids, the concentrations of citric and hydrochloric acids in solution are less than the concentration of acetic acid, a removal of any portion of the hydrogen ions present, through neutralization of basic constituents produced by heat or combination with constituents already present, would affect the acetic acid brine to a less extent than the citric or hydrochloric acid brines.

Besides the direct buffer action of constituents of the vegetable tissues, there are other possible explanations for the observed phenomena of increase of pH value of the acid brines during processing. All or part of the observed change may be due to the neutralization by, or chemical combination or loose conjugation of a part of the acid added with, basic constituents that either are naturally present or are elaborated during the heating process. Or some of the effect noted might be due to the relatively simple phenomena of dilution.

*Amount of Change in pH after Processing, Third Season's Tests.*—Since definite conclusions as to the cause of the observed changes in pH values could not be drawn from the results of the first two seasons' tests, the samples during the third season were prepared for the purpose of

determining whether loss of acid through neutralization, chemical combination, or adsorption might not play a rôle in the observed phenomena. As previously indicated, the samples were carefully prepared in order that an accurate acid balance might be undertaken. The ingoing weight of vegetable and brine, the total acid content, and the pH value of the vegetables and the brines used in the test, and the specific gravity of the brines were determined before canning (table 8). Total acid was deter-

TABLE 8

TOTAL ACID CONCENTRATION AND pH VALUE OF BRINES USED IN CANNING PEAS AND STRING BEANS, THIRD SEASON'S TESTS

| Acetic acid | | Citric acid | | Hydrochloric acid | |
|---|---|---|---|---|---|
| Grams per 100 cc | pH | Grams per 100 cc | pH | Grams per 100 cc | pH |
| 0.103 | 3.18 | 0.103 | 2.69 | 0.103 | 1.58 |
| 0.205 | 3.02 | 0.202 | 2.50 | 0.204 | 1.29 |
| 0.404 | 2.87 | 0.400 | 2.33 | 0.415 | 0.98 |
| 0.762 | 2.72 | 0.754 | 2.16 | 0.765 | 0.73 |
| 1.250 | 2.61 | 1.257 | 2.04 | 1.277 | 0.50 |
| 2.000 | 2.50 | 2.010 | 1.92 | 2.038 | 0.30 |

mined on the brines and vegetable juices by direct titration and by the indirect-titration method of Hartmann and Hillig (*8*). The pH values of the brines and vegetable juices were determined by means of the quinhydrone electrode.

After storage for six months at 0° C, duplicate sets of cans were removed from storage and allowed to reach room temperature. The drained weights of the vegetables were determined by the usual procedure, after which pH values, total acid concentrations, and specific gravity of the drained brine were determined by the methods previously used.

Analysis of the pea and string-bean juices gave pH values of 6.50 and 5.70 respectively. The pea juice required 26.0 cc 0.1 *N* NaOH and the string-bean juice 31.0 cc 0.1 *N* NaOH per 100 grams to neutralize the acids present. The above values are calculated from those obtained by direct titration of 10-gram samples. There was little difference between the results of the direct titrations and those obtained by the indirect method of Hartmann and Hillig. All the following calculations are based upon the results obtained by direct titration.

The pH value of the brines drained from the peas and string beans after canning and six months' storage are recorded in table 9. They are in close agreement with those obtained during the preceding two seasons'

TABLE 9

pH Value of the Brines Drained from String Beans and Peas After Canning in Brines Acidified with Acetic, Citric, and Hydrochloric Acids

| Total acid per 100 cc of brine | Acetic acid | | Citric acid | | Hydrochloric acid | |
|---|---|---|---|---|---|---|
| | Peas | String beans | Peas | String beans | Peas | String beans |
| *grams* | *pH* | *pH* | *pH* | *pH* | *pH* | *pH* |
| 0.0 | 6.40 | 5.52 | 6.40 | 5.52 | 6.40 | 5.52 |
| 0.1 | 6.11 | 5.19 | 6.13 | 5.21 | 5.79 | 4.73 |
| 0.2 | 5.72 | 4.86 | 5.80 | 4.76 | 5.14 | 3.95 |
| 0.4 | 5.21 | 4.56 | 5.26 | 4.38 | 4.32 | 2.91 |
| 0.75 | 4.78 | 4.29 | 4.73 | 4.02 | 3.21 | 1.98 |
| 1.25 | 4.50 | 4.10 | 4.32 | 3.64 | 2.16 | 1.43 |
| 2.0 | 4.28 | 3.93 | 3.94 | 3.33 | 1.23 | 1.10 |

TABLE 10

Calculation of Acid Balances in Peas Canned in Acid Brines

| Brine | 0.1 *N* NaOH required to neutralize acid in product | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Before canning | | | After canning | | | Difference ($A-B$) | Per cent acid lost during canning |
| | In vegetable | In brine | Total ($A$) | In vegetable | In brine | Total ($B$) | | |
| | *cc* | *cc* | *cc* | *cc* | *cc* | *cc* | *cc* | *per cent* |
| Nonacidified | 55.4 | 00.0 | 55.4 | 35.0 | 16.7 | 51.8 | 3.6 | 6.5 |
| Acetic acid brine | 55.5 | 15.8 | 71.3 | 33.4 | 18.4 | 51.8 | 19.5 | 27.3 |
| | 55.4 | 30.6 | 86.0 | 37.9 | 20.8 | 58.7 | 27.3 | 31.7 |
| | 55.5 | 64.3 | 119.8 | 48.7 | 29.8 | 78.5 | 41.3 | 34.5 |
| | 55.4 | 121.8 | 177.2 | 78.0 | 48.1 | 126.1 | 51.1 | 28.8 |
| | 55.5 | 202.0 | 257.5 | 121.8 | 78.2 | 200.0 | 57.5 | 22.3 |
| | 55.4 | 316.0 | 371.4 | 196.0 | 122.8 | 318.8 | 52.6 | 14.2 |
| Citric acid brine | 55.4 | 15.0 | 70.4 | 29.4 | 16.6 | 46.0 | 24.4 | 34.7 |
| | 55.4 | 27.8 | 83.2 | 33.2 | 17.9 | 51.1 | 32.1 | 38.7 |
| | 55.4 | 54.0 | 109.0 | 48.7 | 25.5 | 74.2 | 34.8 | 31.9 |
| | 55.4 | 109.0 | 164.4 | 67.5 | 37.2 | 104.7 | 59.7 | 36.3 |
| | 55.4 | 178.0 | 233.4 | 107.0 | 59.9 | 166.9 | 66.5 | 28.5 |
| | 55.5 | 286.0 | 341.5 | 175.0 | 94.3 | 269.3 | 72.2 | 21.1 |
| Hydrochloric acid brine | 55.4 | 27.3 | 82.7 | 34.8 | 20.9 | 55.7 | 27.0 | 32.6 |
| | 55.3 | 48.2 | 103.5 | 40.7 | 20.2 | 60.9 | 42.6 | 41.2 |
| | 55.3 | 84.2 | 139.5 | 58.9 | 21.8 | 80.7 | 58.8 | 42.2 |
| | 55.2 | 153.1 | 208.3 | 98.1 | 35.5 | 133.6 | 74.7 | 35.9 |
| | 55.4 | 214.8 | 270.2 | 192.8 | 66.3 | 259.1 | 11.1 | 4.1 |
| | 55.3 | 388.8 | 444.1 | 321.8 | 139.0 | 460.8 | −16.7 | −3.8 |

tests but are included because they show the buffering effect of a similar lot of vegetable material towards the three acids.

*Changes in Total Acidity.*—From the data obtained the third season, acid balances were calculated. The results of these calculations are recorded in tables 10 and 11 and are the averages of duplicate samples in all cases.

An examination of tables 10 and 11 reveals that no significant change in total acid content occurs during the canning of peas or string beans in nonacidified brines, although significant changes did occur in pH value (table 9). The deviations in results obtained in calculating the acid balance are within the limits of experimental error.

TABLE 11

CALCULATION OF ACID BALANCES IN STRING BEANS CANNED IN ACID BRINES

| Brine | 0.1 *N* NaOH required to neutralize acid in product | | | | | | | Per cent acid lost during canning |
|---|---|---|---|---|---|---|---|---|
| | Before canning | | | After canning | | | Difference (*A*−*B*) | |
| | In vegetable | In brine | Total (*A*) | In vegetable | In brine | Total (*B*) | | |
| | *cc* | *cc* | *cc* | *cc* | *cc* | *cc* | *cc* | *per cent* |
| Nonacidified | 44.4 | 00.0 | 44.4 | 26.2 | 17.6 | 43.8 | 0.6 | 1.4 |
| Acetic acid brine | 44.6 | 17.0 | 61.6 | 24.7 | 15.9 | 40.6 | 21.0 | 34.1 |
| | 44.4 | 34.4 | 78.8 | 31.1 | 20.7 | 51.8 | 27.0 | 34.3 |
| | 44.6 | 65.8 | 110.4 | 48.5 | 30.9 | 79.4 | 30.8 | 28.0 |
| | 45.3 | 126.8 | 172.1 | 87.0 | 56.2 | 143.2 | 28.9 | 16.8 |
| | 43.5 | 207.0 | 250.5 | 129.8 | 86.8 | 216.6 | 33.9 | 13.5 |
| | 44.3 | 327.0 | 371.3 | 207.0 | 129.0 | 336.0 | 35.3 | 9.5 |
| Citric acid brine | 44.6 | 16.1 | 60.7 | 24.6 | 16.3 | 40.9 | 19.8 | 32.6 |
| | 44.6 | 31.5 | 76.1 | 31.0 | 19.6 | 50.6 | 25.5 | 33.5 |
| | 44.6 | 61.0 | 105.6 | 46.1 | 28.9 | 75.0 | 30.6 | 29.0 |
| | 44.6 | 109.8 | 154.4 | 75.2 | 45.0 | 120.2 | 34.2 | 21.2 |
| | 44.3 | 193.0 | 237.3 | 118.1 | 74.5 | 192.6 | 44.7 | 18.8 |
| | 44.5 | 306.0 | 350.5 | 179.0 | 120.0 | 299.0 | 51.5 | 14.7 |
| Hydrochloric acid brine | 44.5 | 25.6 | 70.1 | 27.8 | 16.6 | 44.4 | 25.7 | 36.7 |
| | 44.5 | 53.0 | 97.5 | 37.2 | 23.0 | 60.2 | 37.3 | 38.3 |
| | 44.4 | 111.2 | 155.6 | 64.7 | 41.6 | 106.3 | 49.3 | 31.7 |
| | 44.4 | 202.0 | 246.4 | 118.0 | 80.2 | 198.2 | 48.2 | 19.6 |
| | 44.5 | 336.0 | 380.5 | 210.0 | 131.0 | 341.0 | 39.5 | 10.4 |
| | 44.5 | 548.0 | 592.5 | 360.0 | 194.5 | 554.5 | 38.0 | 6.4 |

On the other hand, rather large losses of total acid occur when these vegetables are canned in brines acidified with acetic, citric, and the lower concentrations of hydrochloric acids. The loss of acid increases with the concentration of acid added to the product, except in those samples canned in high concentrations of hydrochloric acid brine. In the pea samples canned in brine containing 1.25 grams of hydrochloric acid per 100 cc, little or no acid is lost, while in those canned with brine containing 2.0 grams of hydrochloric acid per 100 cc, there is evidence that a production of acid occurs. In general, the loss of acid is greater in canned peas than in canned string beans.

Loss of acid therefore seems to account for part of the increase in pH value that occurs when vegetables are canned in acidified brines. Al-

though the loss of acid is insufficient to account for the entire change, it nevertheless contributes to the final result. The results of this experiment, however, do not indicate whether the acid was used in neutralization of some basic constituents, or in decomposition of some of the more complex constituents, or whether it is bound by colloidal constituents present in the vegetable tissues.

TABLE 12

Total Acidity and pH Relations of Acidified Pea Juice, before and after Heating

| Brine | Milliequivalents of acid present in original sample* | Milliequivalents of acid recovered | | Difference | | pH value | |
|---|---|---|---|---|---|---|---|
| | | Before heating | After heating | Before heating (col. 1—col. 2) | After heating (col. 1—col. 3) | Before heating | After heating |
| | *1* | *2* | *3* | *4* | *5* | *6* | *7* |
| Control ($H_2O$) | 0.650 | 0.643 | 0.540 | 0.007 | 0.110 | 6.60 | 6.72 |
| Acetic acid brine | 0.997 | 0.855 | 0.675 | 0.142 | 0.322 | 6.33 | 6.29 |
| | 1.330 | 0.878 | 0.882 | 0.452 | 0.448 | 6.00 | 5.93 |
| | 1.991 | 1.440 | 1.372 | 0.551 | 0.619 | 5.28 | 5.31 |
| | 3.179 | 2.520 | 2.564 | 0.659 | 0.615 | 4.76 | 4.78 |
| | 4.794 | 4.095 | 3.982 | 0.699 | 0.812 | 4.47 | 4.46 |
| | 7.310 | 6.638 | 6.435 | 0.672 | 0.875 | 4.23 | 4.22 |
| Citric acid brine | 0.979 | 0.890 | 0.585 | 0.089 | 0.394 | 6.33 | 6.39 |
| | 1.280 | 1.056 | 0.810 | 0.224 | 0.470 | 6.11 | 6.17 |
| | 1.888 | 1.373 | 1.283 | 0.515 | 0.605 | 5.36 | 5.43 |
| | 2.989 | 2.349 | 2.204 | 0.640 | 0.785 | 4.68 | 4.74 |
| | 4.570 | 3.780 | 3.622 | 0.790 | 0.948 | 4.25 | 4.27 |
| | 6.950 | 5.985 | 5.875 | 0.965 | 1.075 | 3.87 | 3.86 |
| Hydrochloric acid brine | 1.213 | 0.945 | 0.810 | 0.268 | 0.403 | 5.95 | 6.07 |
| | 1.775 | 1.598 | 1.620 | 0.177 | 0.155 | 5.30 | 5.37 |
| | 2.930 | 2.399 | 1.980 | 0.531 | 0.950 | 4.24 | 4.22 |
| | 4.860 | 3.713 | 3.726 | 1.147 | 1.134 | 2.99 | 2.98 |
| | 7.692 | 6.480 | 6.772 | 1.212 | 0.920 | 1.84 | 1.90 |

* That present in pea juice plus that added.

## TOTAL ACIDITY OF EXPRESSED JUICES MIXED WITH ACIDIFIED BRINES

Since the experiment just described yielded little evidence concerning the mechanism of the reactions involved, largely because the changes occurring during the canning operations cannot be observed, a series of samples were prepared using the juices obtained from the same lot of vegetables, in which the ratio of vegetable to brine was approximately the same as in the canning tests. The series consisted of 25-cc aliquots of pea and string-bean juice plus 20-cc portions of the acidified brines used

in the canning tests. Total acidity was determined by direct titration and pH values by the quinhydrone electrode on each sample before and after heating for 1 hour at 100° C. The results of this experiment are given in tables 12 and 13.

When an aliquot of pea juice is acidified (table 12), stirred, and allowed to stand for some minutes, subsequent titration does not account for all the acid added plus that present in the juice itself. A portion of

TABLE 13

TOTAL ACIDITY AND pH RELATIONS OF ACIDIFIED STRING-BEAN JUICE BEFORE AND AFTER HEATING

| Brine | Milliequivalents of acid present in original sample* | Milliequivalents of acid recovered | | Difference | | pH value | |
|---|---|---|---|---|---|---|---|
| | | Before heating | After heating | Before heating (col. 1—col. 2) | After heating (col. 1—col. 3) | Before heating | After heating |
| | *1* | *2* | *3* | *4* | *5* | *6* | *7* |
| Control ($H_2O$)..... | 0.825 | 0.833 | 1.058 | −0.008 | −0.233 | 5.74 | 5.62 |
| Acetic acid brine..... | 1.171 | 1.237 | 1.270 | −0.066 | −0.099 | 4.98 | 5.01 |
| | 1.504 | 1.530 | 1.570 | −0.026 | −0.066 | 4.72 | 4.75 |
| | 2.121 | 2.160 | 2.178 | −0.039 | −0.057 | 4.42 | 4.47 |
| | 3.354 | 3.352 | 3.398 | 0.002 | −0.044 | 4.16 | 4.21 |
| | 4.968 | 4.950 | 4.975 | 0.018 | −0.007 | 3.96 | 4.03 |
| | 7.485 | 7.422 | 7.460 | 0.063 | 0.025 | 3.79 | 3.86 |
| Citric acid brine..... | 1.153 | 1.170 | 1.170 | −0.017 | −0.017 | 4.99 | 5.00 |
| | 1.455 | 1.462 | 1.463 | −0.007 | −0.008 | 4.65 | 4.67 |
| | 2.062 | 2.057 | 2.048 | 0.005 | 0.014 | 4.26 | 4.30 |
| | 3.168 | 3.082 | 3.117 | 0.086 | 0.051 | 3.86 | 3.95 |
| | 4.745 | 4.648 | 4.680 | 0.097 | 0.065 | 3.52 | 3.60 |
| | 7.125 | 6.975 | 7.085 | 0.150 | 0.040 | 3.20 | 3.30 |
| Hydrochloric acid brine..... | 1.328 | 1.417 | 1.440 | −0.089 | −0.112 | 4.55 | 4.59 |
| | 1.950 | 1.845 | 1.935 | 0.105 | 0.015 | 3.83 | 3.90 |
| | 3.103 | 2.835 | 2.993 | 0.268 | 0.110 | 2.80 | 2.92 |
| | 5.033 | 4.838 | 4.972 | 0.195 | 0.061 | 1.88 | 1.94 |
| | 7.865 | 7.560 | 7.875 | 0.305 | −0.010 | 1.24 | 1.25 |
| | 12.125 | 11.750 | 12.280 | 0.375 | −0.155 | 0.87 | 0.86 |

* That present in string-bean juice plus that added.

the acid evidently becomes so bound that it no longer furnishes hydrogen ions detectable by titration. The extent of the reaction is dependent upon the kind and amount of acid added, being greater the higher the concentration and the stronger the acid added. Although the reaction by means of which the acid is fixed occurs in the cold, it is increased in extent by heating except in the samples containing high concentration of hydrochloric acid, where the data would indicate that a liberation of part of the bound acid occurs during heating.

String-bean juice (table 13) behaves somewhat differently from pea juice when treated with acidified brines. When acetic acid brines containing up to 0.4 gram per 100 cc, citric acid brines containing up to 0.2 gram per 100 cc of acid, or hydrochloric acid brine containing 0.1 gram per 100 cc, are added to the juice subsequent titration indicates more acid than the total of that originally added plus the amount present in the vegetable tissue. When brines of higher acidity than those mentioned are added, less acid is found than was originally present, and the amount of acid lost depends upon the kind and concentration of the acid added, as was previously found for pea juice. The data in table 13 show that the increase in total acid occurred only in those samples in which the amount of acid added was insufficient to lower the pH below 4.42. Apparently string beans contain a colloidal system having an isoelectric point of approximately pH 4.3, which is capable of binding a portion of the acid added in excess of the amount necessary to lower the pH beyond the isoelectric point. This colloidal system is precipitated by hydrogen ions.

Heating the samples resulted in an increase in total acid in nearly all the samples over the amounts found in the unheated samples (table 13). This increase in acid was greatest in the samples acidified with hydrochloric acid brines, in which it was large enough at the higher acid concentrations to account for all the acid originally bound and to increase the total acid above that originally present. The increase in total acid as a result of the heating was accompanied by a slight increase in pH value of the samples.

The behavior of the samples of pea juice during preparation indicates that the results obtained were due largely to changes induced in the colloidal systems of the juices by the added acids. The addition of acid to pea juice first causes some of the suspended material to settle out, and leaves the supernatant liquid light green and cloudy. Further addition causes the liquid to become colorless but to remain cloudy and the suspended material to become grayish green in color. The next change is a coagulation and complete flocculation of all suspended material, upon which the liquid becomes colorless and clear. Further addition of acid causes color changes in both the suspended matter and the liquid, with the latter increasing in cloudiness as the acid is increased.

These changes occur at definite pH values and are not functions of the kind of acid used. The change of the liquid from green to colorless occurs at a pH value of 5.3 in the pea juice, and coagulation and clearing of the liquid occurs at approximately 4.5.

Heating caused the suspended material in all the pea-juice samples to

coagulate, except those to which the three highest concentrations of hydrochloric acid brine were added. These samples were cloudy and rather viscous. The liquid in all the samples turned brown upon heating and was brilliantly clear. The coagulated sediment was yellowish green in color.

The changes induced by acid added to string-bean juice are likewise a function of the pH value. A very small amount of acid added to string-

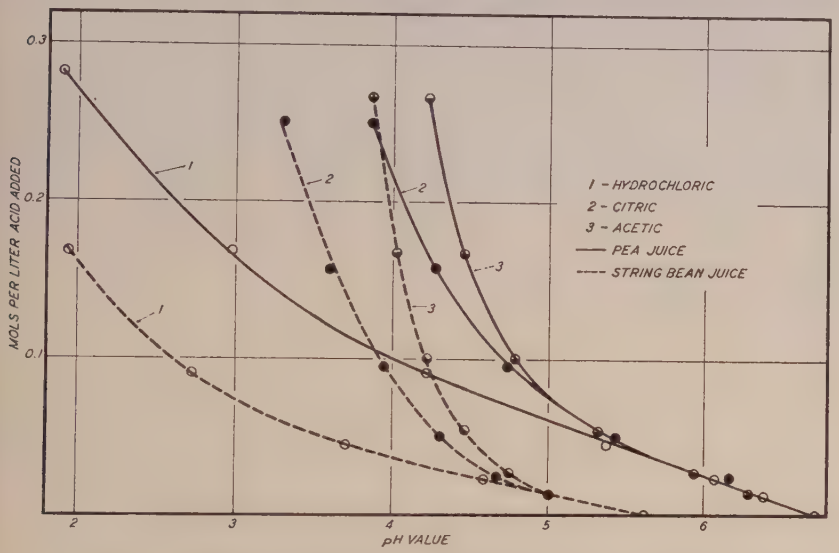


**Fig. 7.—The buffering action of string-bean and pea juice toward hydrochloric, citric, and acetic acid.**

bean juice resulted in some coagulation of the suspended material. However, the liquid did not become clear until its pH value was lowered to 4.7, and it reclouded at pH 3.5. The changes brought about by heat were similar in all respects to those already recorded for pea juice.

The difference in buffer capacity of the respective vegetable juices previously noted is therefore partially explained by the nature of the colloidal system present and the degree to which it is affected by the added acid. Peas, which have as strong a buffer capacity as any vegetable tested, owe a large proportion of their ability to resist changes in pH value upon acidification, to the presence of a colloidal system which binds a portion of the acid added. String beans, which are among the vegetables having a low buffer capacity, do not contain a colloidal system comparable to that of peas, and the buffer effect noted is undoubtedly due in large part to the presence of acid-salt systems.

In figure 7 the pH values recorded in this experiment have been plotted against the calculated mols per liter of acid added, for each vegetable and each acid used. The curves show that at low concentrations each acid is buffered to the same extent by the vegetable under examination. At higher concentrations, the buffering effect of the acid itself becomes apparent and is responsible for the differences noted. The Van Slyke B value for peas at pH 6 taken from this graph is 0.034 and the string beans at pH 5 is 0.022. These values are in agreement with the previously reported values for these two vegetable juices.

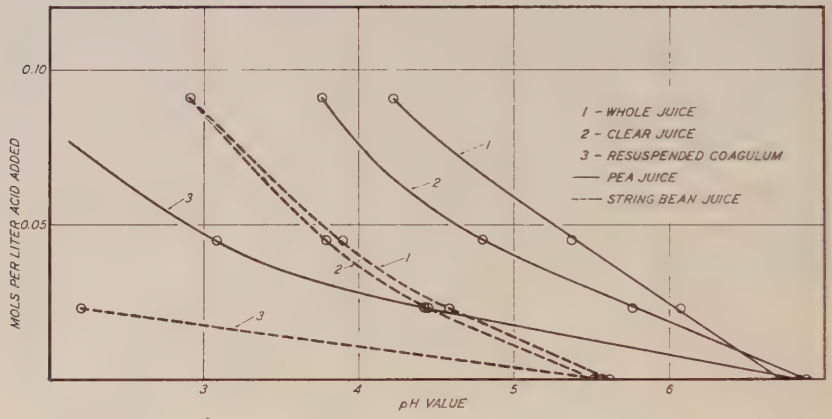


Fig. 8.—The buffering action of various fractions of string-bean and pea juice toward hydrochloric acid.

## BUFFER CAPACITY OF HEAT-PRECIPITABLE SUBSTANCES

As previously stated, heating causes a coagulation of suspended matter in both these juices, which readily flocculates and leaves the remaining liquor brilliantly clear. There was evidence that these heat-precipitable substances played a rôle in the capacity of the juice to act as buffers.

Portions of both juices were prepared from the previously ground and frozen material, placed in tightly sealed containers and pasteurized for 1 hour at 100° C. The coagulum was filtered out and the filtrate obtained was set aside. The coagulum was then washed and was resuspended in a volume of distilled water equivalent to the quantity of juice from which it was obtained. Aliquot portions of both the filtrate and the resuspended coagulum were then treated with aliquot portions of the acidified brines used in the previous experiments in the ratio of 25 cc of juice to 20 cc of brine and pH determinations were made upon each sample. Only the data obtained from the pH determinations made upon the samples acidi-

fied with hydrochloric acid brine are presented in figure 8, since they are illustrative of the points of chief interest.

The changes in pH resulting from the addition of acid to the samples calculated as mols per liter are shown for the unheated whole juice, the clear filtrate, and the resuspended coagulum for both vegetables. The curves show that the clear filtrate from the heated pea juice has much less buffer capacity than the whole juice, and likewise that very little change in buffer capacity has occurred in the string-bean juice. The curves further show that the resuspended coagulum of both peas and string beans has a definite, although somewhat limited buffer capacity.

The Van Slyke B values for the unheated pea juice, the clear filtrate, and the resuspended coagulum from pea juice are 0.034, 0.021, and 0.010 respectively at a pH value of 6.0, and for the same materials from string-bean juice, 0.022, 0.021, and 0.007 at a pH value of 5.0. The filtrate from pea juice, therefore, has a buffer capacity of the same magnitude as the string-bean juice. This fact would indicate that the materials responsible for the buffer capacity of peas in excess of that shown by other vegetable juices are chiefly composed of heat-coagulable substances. These are probably colloidal protein or carbohydrate. The heat-coagulable substance present in string-bean juice has no influence upon the buffer capacity of the juice, although it does exert a small but definite buffering action when free of juice. The chemical composition of these substances was not determined.

That dilution might account for part of the increase in pH value of the acidified brines has been previously suggested. A considerable dilution does occur when the contents of the cans are caused to mix intimately by heating. The 150-gram portions of vegetable material used contain approximately 125 cc of water which mixes with the 100-cc portions of the acidified brines. However, when 25-cc quantities of water were mixed with 20-cc portions of the acidified brines tested, the change in pH value that occurred was only 0.40 pH unit for hydrochloric acid, 0.24 pH unit for citric acid, and 0.20 pH unit for acetic acid. Dilution is therefore not an important factor in the phenomena observed.

## CONCLUSIONS

The buffer capacity of the expressed juices of the vegetables tested was effected to only a slight extent by heat when the juices were heated in contact with acid but marked changes occurred when the juices were heated in contact with base. Heating prior to the addition of acid causes a marked change in the buffer capacity of pea juice as determined by potentiometric titration with base.

Peas and artichokes have the highest buffer indexes of the various vegetables tested, followed in order by green asparagus, string beans, and white asparagus.

Acidified brines increase markedly in pH value during canning with nonacid vegetables. The change in pH value is less with acetic acid brines than with citric acid brines of the same low initial total acid concentration. With brines of the same initial pH value markedly less change occurs in citric acid brines than in acetic acid brines. Peas exert a greater buffering effect on acidified brines than any of the other vegetables tested.

A definite loss of acid occurs when vegetables are canned with acidified brines which accounts for a part, at least, of the increase in pH values noted. Probably the acid lost is involved in adsorption reactions with the colloidal systems of the vegetable tissues. The total buffer effect noted, however, cannot be ascribed to any single substance or system but is complex in nature.

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